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Bioherbicidal Properties of *Parthenium hysterophorus*, *Cleome rutidosperma* and *Borreria alata* Extracts on Selected Crop and Weed Species

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Abstract: Natural product-based herbicides could be the effective alternatives to synthetic chemical herbicides for eco-friendly weed management. This research, therefore, was conducted to identify the phytotoxic properties of *Parthenium hysterophorus* L., *Cleome rutidosperma* DC. and *Borreria alata* (Aubl.) DC. with a view to introducing them as a tool for natural herbicide development. The methanol extracts of these plants were examined on the germination and growth of *Zea mays* L., *Oryza sativa* L., *Abelmoschus esculentus* (L.) Moench and *Amaranthus gangeticus* L., *Oryza sativa* f. *Spontanea* Roshev. (Weedy rice), *Echinochloa colona* (L.) Link., *Euphorbia hirta* L., and *Ageratum conyzoides* L. under laboratory and glasshouse conditions. A complete randomized design (CRD) with five replications and randomized complete block design (RCBD) with four replications were laid out for laboratory and glasshouse experiments, respectively. In the laboratory experiment, three plant extracts of 0, 6.25, 12.5, 50, and 100 g L^{-1} were tested on survival rate, hypocotyl, and radicle length of eight test plant species. No seed germination of *A. conzyoides*, *E. hirta*, and *A. gangeticus* were recorded when *P. hysterophorus* extract was applied at 50 g L^{-1}. *C. rutidosperma* had the same effect on those plants at 100 g L^{-1}. In the glasshouse, similar extracts and concentrations used in the laboratory experiments were sprayed on at the 2–3 leaf stage for grasses and 4–6 for the broadleaf species. Tested plants were less sensitive to *C. rutidosperma* and *B. alata* compared to *P. hysterophorus* extract. Among the weeds and crops, *A. conyzoides*, *E. hirta*, *A. esculentus* and *A. gangeticus* were mostly inhibited by *P. hysterophorus* extract at 100 g L^{-1}. Based on these results, *P. hysterophorus* was the most phytotoxic among the tested plant extracts and could be used for developing a new natural herbicide for green agriculture.

Keywords: allelopathy; bioherbicide; *P. hysterophorus*; germination; growth

1. Introduction

Parthenium hysterophorus L., *Cleome rutidosperma* DC., and *Borreria alata* (Aubl.) DC. belong to the family Asteraceae, Cleomaceae, and Rubiaceae, respectively, and are invasive weeds in Malaysia. Their damage includes threats to biodiversity, exacerbation of allergies and dermatitis, mutagenesis in humans and livestock, and interference (competition and allelopathy) with field crops and rangelands [1–3]. *Parthenium hysterophorus* is native to Mexico and has been spreading like wildfire in different countries. At present, ten states of Malaysia are invaded by this weed, and the state Kedah is the worst infested area [4]. Yield losses between 21 to 50% have occurred due to *P. hysterophorus* in different crop fields [5]. The allelochemicals released from this plant may inhibit seedling growth and nutrient uptake by destroying the plants' usable source of nutrients [6,7]. *Cleome rutidosperma* is native to tropical Africa and now naturalized in different regions of Asia, Australia, America, and the West Indies [2]. This weed is mainly found in sugarcane fields and causes more than 50% sugarcane yield reduction in the Philippines [2]. In Malaysia, *C. rutidosperma* is planted around the field edges by the farmers to divert the ovipositor of *Plutella xylostella* (diamondback moth) away from the crop field [2]. Moreover, the plant is considered as the alternate host of *Meloidogyne javanica* and *Meloidogyne incognita* [8]. *Borreria alata* is *native* to southern Mexico [9]. It is a common weed in sugarcane, rubber, oil palm, orchards, tea, chinchoa, cassava, and many annual upland crops such as maize, soybean, and rice. It has reduced the dry weight and height of young rubber by 12 and 17%, respectively [10].

While manual weed control is the best and most sustainable, herbicidal control is the more effective and rapid option for weed management. However, due to the migration of labor from agriculture to industry; and off-target toxicity including weed biotypes resistant to existing synthetic herbicides, researchers are motivated to think about alternatives [11]. In these circumstances, allelopathic plants or their isolated allelochemicals may play a key role. Allelopathic plants may exert either inhibitory or stimulatory effects on the germination and growth of other plants in their immediate vicinity. Recently, allelopathic plants or their allelochemicals are being utilized sporadically instead of synthetic herbicides to control weeds, and more attention has been paid by scientists to develop natural product-based herbicides from allelopathic plants [12–16]. Moreover, due to the presence of higher oxygen and nitrogen-rich molecules and having a relatively low halogen substitute, most of the allelochemicals are environment friendly [17]. Hence, the allelochemicals isolated from allelopathic plants can either be developed as a natural herbicide or used for templates to further develop novel synthetic herbicides with new modes of action [14,16].

In most of the allelopathic studies, laboratory bioassay has mainly been used by the researchers to identify allelopathic plants because of their rapid out return. Additionally, bioassay performance may help the researchers to predict the allelopathic potentials of a plant in glasshouse or field conditions [18]. However, a plant that showed strong allelopathic activity on target plants in s laboratory bioassay, might differ in glasshouse or field conditions due to the influence of several environmental determinants [19]. Plant crude extract with strong allelopathic potential has recently contributed greatly to weed management and controlling weeds [19–22]. Although the allelopathic properties of *P. hysterophorus* have been reported, the allelopathy of *C. rutidosperma* and *B. alata* are scant so far. In addition, most of the articles related to *P. hysterophorus* allelopathy were based on their laboratory bioassay results. Therefore, a detailed study of these plants under both laboratory and glasshouse conditions warrants due attention to evaluating their allelopathic potential. Hence, to explore the allelopathic properties of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* on *Zea mays* L., *Oryza sativa* L., *Abelmoschus esculentus* (L.) Moench, *Amaranthus gangeticus* L., *Oryza sativa* f. *spontanea* Roshev (Weedy rice), *Echinochloa colona* (L.) Link., *Euphorbia hirta* L. and *Ageratum conyzoides* L., two experiments were conducted under laboratory and glasshouse conditions with a view to developing natural product-based bioherbicides.

2. Materials and Methods

2.1. Test Plants

Four cropspecies, e.g., *Z. mays*, *O. sativa*, *A. esculentus*, and *A. gangeticus*, and four weed species, e.g., Weedy rice, *E. colona*, *E. hirta*, and *A. conzyoides* were used in this research as test plants. The seeds of *E. colona*, Weedy rice, and *O. sativa* "MR 219" were collected from the rice fields of Sekinchan, Kuala Selangor, and Selangor, Malaysia, and other weed seeds (*E. hirta* and *A. conzyoides*) were collected from farm 15 at the Universiti Putra Malaysia.

The seeds of *Z. mays*, *A. esculentus*, and *A. gangeticus* were purchased from Green World Genetics Sdn. Bhd., Kuala lumpur, Selangor, Malaysia.

2.2. Extraction Procedure

Whole plants of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* at their maximum vegetative stage were collected from different locations of Universiti Putra Malaysia except *P. hysterophorus* which was collected from Ladang Infoternak, Sungai Siput, Perak, Malaysia. The collected weeds were washed carefully with running tap water for removing the dust particles, then air-dried in open trays under shaded conditions at room temperature for 3 weeks. Then, each species was chopped and crushed separately in a Willey mill. In a conical flask, 100 g powder of each species was soaked with 1000 mL methanol: distilled water (80:20, v/v), and paraffin was used for wrapping the flask. The flask was shaken using an Orbital shaker at 150 rpm agitation speed for 48 h at a room temperature (24–26 °C). The solution was filtered using four layers of cheesecloth then centrifuged for 1 h at 3000 rpm. Then a 0.2-μ, 15-mm syringe filter (Phenex, Non-sterile, Luer/Slip, LT Resources, Puchong, Selangor, Malaysia) was used to re-filter the solution. A rotary evaporator was used at 40 °C to evaporate the methanol from the extract. Sterile distilled water was used to obtain stock extract concentrations of 6.25, 12.5, 25, 50, and 100 g L^{-1} for bioassay. All extracts were refrigerated at 4 °C before they were used. The method described by Aslani et al. [23] was used in preparing methanol extracts for each species.

2.3. Laboratory Bioassay

2.3.1. Experimental Site

The experiment was conducted in a growth chamber at the Seed Technology Laboratory, Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia (3°02′ N, 101°42′ E, 31 m elevation), Selangor, Malaysia from January to March 2019.

2.3.2. Experimental Treatments and Design

Healthy and uniform weed seeds were collected and then soaked for 24 h with 0.2% potassium nitrate (KNO_3) and then rinsed with distilled water and incubated at room (24–26 °C) temperature until the radicle emerged about 1 mm. Twenty pre-germinated uniform seeds of each crop and weed species were placed in 9.0-cm-diameter plastic disposable Petri dishes, containing two sheets of Whatman No. 1 filter paper. Then the filter paper on the Petri dishes was moistened with 10 mL of *P. hysterophorus*, *C. rutidosperma* and *B. alata* methanol extract at six different concentrations: 0 (distilled water only), 6.25, 12.5, 25, 50, and 100 g L^{-1} separately. The Petri dishes were then moved to a growth chamber and incubated under fluorescent light (8500 lux) with a completely randomized design at the temperature of 30/20 °C (day/night), with a 12 h/12 h (day/night cycle). Relative humidity ranged from 30 to 50%. The lids of the Petri dishes were not sealed to allow gas exchange and to avoid an anaerobic condition.

2.3.3. Data Collection

After 7 days of germination, the survival rate, radicle and hypocotyl length of all seedlings were measured. The hypocotyl and radicle length was measured using Image J software [24] and the inhibitory effect was calculated by the equation stated below [23]

$$I = 100\,(C - A)/C$$

where "I" is the inhibition amount (%), "C" is the mean length of the radicle and hypocotyl of the control, and "A" is the mean length of the radicle and hypocotyl of the methanol extracts.

2.4. Glasshouse Experiment

2.4.1. Experimental Site and Design

The glasshouse experiment was conducted from April to June 2020, at the faculty of Agriculture in Ladang 15, Universiti Putra Malaysia. Foliar application of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* methanol extract was assessed for their suppressive effects on the growth and development of Weedy rice, *E. colona*, *E. hirta*, *A. conyzoides*, *Z. mays*, *O. sativa*, *A. esculentus*, and *A. gangeticus*. Pre-germinated seeds of test plants were sown in each pot (15 cm diameter × 12 cm height) and then covered with soil at a depth of 1 cm, and finally, the soil was moistened with tap water. After germination, only five equal-sized healthy seedlings of *O. sativa*, *E. colona*, *E. hirta*, *A. conyzoides*, and weedy rice, and one seedling (equal-sized healthy) of *Z. mays*, *A. gangeticus*, and *A. tricolor* were kept in each pot. The pots were arranged in a randomized complete block design with four replications. Methanol extracts of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* were sprayed with 6.25, 12.5, 25, 50, and 100 g L^{-1} concentration at the 2–3 leaf stage (2 weeks old) for grasses and 4–6 leaf stage for broadleaf species (3 weeks old) with the help of a 1 L multipurpose sprayer (Deluxe pressure sprayer). Spray volume (100 mL m^{-2}) was prepared using distilled water [25]. Plants in the control treatment were sprayed with 200 mL water without extract at two days intervals or when needed.

2.4.2. Data Collection

Three weeks after spray, the individual plant was separated into the root, shoot, and leaf fractions. Plant height (PH) and root length (RL) were measured using a 1 m ruler. The leaf area was determined using a leaf area meter (LI-3000, Li-COR, Lincoln, NE, USA) and expressed as cm^2 $plant^{-1}$. Fresh and dry weights were determined using a digital balance. Samples were dried in an oven at 60 °C for 72 h to take the dry weight of the samples. Total chlorophyll content indicated as SPAD value was measured by a chlorophyll meter, SPAD-502 (Menolta, Japan), as described by Mahdavikia et al. [26]

2.5. Statistical Analysis

A two-way analysis of variance (ANOVA) was performed to determine any significant differences between each treatment and the control for both experiments. The differences between the treatment's means were pooled using the Tukey test with a 0.05 probability level. SAS (Statistical Analysis System) software, version 9.4 (Cary, NC, USA) was used to conduct the analysis. Probit analysis based on the percentage of inhibition of survival rate or radicle and hypocotyl length was used to measure ECs50, ECr50, and ECh50. ECs50, ECr50, and ECh50 were the effective doses capable of inhibiting 50% of survival rate, radicle, and hypocotyl length respectively. The most active extracts were determined as the index (Re) using the equation given below for each extract tested:

$$\text{Rank (Re)} = \text{ECs50n (survival rate)} + \text{ECr50n (radicle)} + \text{ECh50n (hypocotyl)}$$

where Re is the rank of the extract and ECs50n, ECr50n, and ECh50n are the concentrations of treatments that cause 50% inhibition on germination, root, and hypocotyl growth, respectively. The extract with the lowest Re values was considered as the most phytotoxic treatment and the least phytotoxic effect of the extract was observed for the highest Re value.

3. Results

3.1. Laboratory Experiment

3.1.1. Effect of Methanol Extracts on Survival Rate and Initial Growth of Weeds

The results showed that *P. hysterophorus*, *C. rutidosperma*, and *B. alata* extracts significantly influenced the survival rate as well as the hypocotyl and radicle length of the tested weed species ($p < 0.05$). The magnitude of inhibition increased with an increase in extract concentration (Table 1).

Table 1. Effects of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* methanol extracts on survival rate (%), hypocotyl, and radicle length (cm) of the tested weed species.

Tested Plants	Dose (g L^{-1})	*P. hysterophorus*			*C. rutidosperma*			*B. alata*		
		Survival Rate (%)	**Hypocotyl Length (cm)**	**Radicle Length (cm)**	**Survival Rate (%)**	**Hypocotyl Length (cm)**	**Radicle Length (cm)**	**Survival Rate (%)**	**Hypocotyl Length (cm)**	**Radicle Length (cm)**
A. conyzoides	0.00	100.00 a ± 0 (0)	4.91 a ± 0.03 (0)	2.70 a ± 0.04 (0)	100.00 a ± 0 (0)	4.94 a ± 0.02 (0)	2.72 a ± 0.04 (0)	100.00 a ± 0 (0)	4.99 a ± 0.04 (0)	2.68 a ± 0.03 (0)
	6.25	82.00 b ± 1.22 (18)	2.92 b ± 0.04 (40.53)	0.89 b ± 0.02 (67.04)	99.00 a ± 1 (1)	3.56 b ± 0.07 (27.93)	1.30 b ± 0.02 (52.20)	99.00 a ± 1 (1)	3.99 b ± 0.03 (20.04)	1.33 b ± 0.01 (50.37)
	12.5	38.00 c ± 1.3 (62)	0.86 c ± 0.02 (82.48)	0.34 c ± 0.01 (87.40)	72.00 b ± 2 (28)	2.23 c ± 0.02 (54.86)	0.86 c ± 0.01 (68.38)	83.00 b ± 4.64 (17)	2.96 c ± 0.03 (40.68)	1.05 c ± 0.02 (60.82)
	25	13.00 d ± 1.28 (87)	0.41 d ± 0.01 (91.65)	0.17 d ± 0.01 (93.70)	45.00 c ± 3.53 (55)	0.96 d ± 0.01 (80.57)	0.57 d ± 0.12 (79.04)	42.00 c ± 2.55 (58)	1.88 d ± 0.05 (62.32)	0.82 d ± 0.01 (69.40)
	50	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	18.00 d ± 1.22 (72)	0.62 e ± 0.02 (87.44)	0.29 e ± 0.01 (89.34)	28.00 d ± 2.50 (72)	0.98 e ± 0.04 (80.36)	0.71 e ± 0.02 (73.51)
	100	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 f ± 0 (100)	0.00 f ± 0 (100)	5.00 e ±0.32 (95)	0.44 f ± 0.02 (91.18)	0.23 f ± 0.21 (91.42)
E. hirta	0.00	100.00 a ± 0 (0)	6.32 a ± 0.02 (0)	1.91 a ± 0.01 (0)	100.00 a ± 0 (0)	6.26 a ± 0.02 (0)	1.98 a ± 0.03 (0)	100.00 a ± 0 (0)	6.34 a ± 0.02 (0)	1.89 a ± 0.2 (0)
	6.25	84.00 b ± 1.87 (16)	3.68 b ± 0.01 (41.77)	0.71 b ± 0.03 (62.83)	96.00 a ± 1.87 (4)	4.29 b ± 0.01 (31.47)	1.32 b ± 0.02 (33.33)	98.00 a ± 1.22 (2)	4.94 b ± 0.11 (22.08)	0.95 b ± 1.4 (49.74)
	12.5	39.00 c ± 1.8 (61)	0.97 c ± 0.01 (84.65)	0.33 c ± 0.02 (82.72)	78.00 b ± 2 (22)	3.16 c ± 0.2 (49.52)	0.71 c ± 0.08 (64.14)	82.00 b ± 1.2 (18)	3.75 c ± 0.07 (40.85)	0.81 c ± 0.03 (57.14)
	25	12.00 d ± 1.22 (88)	0.52 d ± 0.1 (91.77)	0.16 d ± 0.01 (91.62)	48.00 c ± 1.22 (52)	0.99 d ± 0.04 (84.19)	0.44 d ± 0.7 (77.78)	45.00 c ± 2.24 (55)	2.16 d ± 0.01 (65.93)	0.59 d ± 1.96 (68.78)
	50	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	20.00 d ± 1.58 (80)	0.71 e ± 0.3 (88.66)	0.29 e ± 1.77 (85.35)	29.00 d ± 1.87 (71)	1.20 e ± 0.04 (81.07)	0.45 e ± 0.02 (76.19)
	100	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 f ± 0 (100)	0.00 f ± 0 (100)	9.00 e ± 1 (91)	0.69 f ± 0.2 (89.12)	0.18 f ± 0.01 (90.48)
Weedy rice	0.00	100.00 a ± 0 (0)	4.89 a ± 0.1 (0)	2.13 a ± 0.08 (0)	100.00 a ± 0 (0)	4.98 a ± 0.08 (0)	2.21 a ± 0.1 (0)	100.00 a ± 0 (0)	4.91 a ± 0.14 (0)	2.02 a ± 0.02 (0)
	6.25	87.00 b ± 2.55 (13)	3.23 b ± 0.06 (34.15)	0.80 b ± 0.03 (62.44)	98.00 a ± 1.22 (2)	3.61 b ± 0.01 (27.51)	1.06 b ± 0.02 (52.04)	100.00 a ± 0 (0)	3.98 b ± 0.04 (18.94)	1.02 b ± 0.03 (49.50)
	12.5	49.00 c ± 1.87 (51)	1.48 c ± 0.02 (69.92)	0.60 c ± 0.02 (71.83)	74.00 b ± 1.87 (26)	2.33 c ± 0.02 (53.21)	0.72 c ± 0.04 (67.42)	81.00 b ± 1.87 (19)	2.75 c ± 0.07 (44.38)	0.96 bc ± 0.02 (52.47)
	25	20.00 d ± 1.58 (80)	0.75 d ± 0.01 (84.66)	0.24 d ± 0.01 (88.73)	44.00 c ± 1 (56)	0.99 d ± 0.03 (80.12)	0.45 d ± 0.02 (79.64)	51.00 c ± 1.8 (49)	2.02 d ± 0.03 (58.86)	0.88 c ± 0.01 (56.43)
	50	7.00 e ± 2 (93)	0.24 e ± 0.06 (95.09)	0.16 de ± 0.13 (92.49)	15.00 d ± 3.53 (85)	0.77 e ± 0.01 (84.54)	0.25 d ± 0.01 (88.69)	23.00 d ± 1.22 (77)	1.14 e ± 0.01 (76.78)	0.33 d ± 0.19 (83.66)
	100	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	7.00 e ± 1.27 (93)	0.46 f ± 0.02 (90.63)	0.11 e ± 0.9 (94.55)

Table 1. *Cont.*

Tested Plants	Dose (g L^{-1})	*P. hysterophorus*			*C. rutidosperma*			*B. alata*		
		Survival Rate (%)	**Hypocotyl Length (cm)**	**Radicle Length (cm)**	**Survival Rate (%)**	**Hypocotyl Length (cm)**	**Radicle Length (cm)**	**Survival Rate (%)**	**Hypocotyl Length (cm)**	**Radicle Length (cm)**
E. colona	0.00	100.00 a ± 0 (0)	5.11 a ± 0.03 (0)	2.95 a ± 1.96 (0)	100.00 a ± 0 (0)	5.00 a ± 0.04 (0)	2.94 a ± 0.04 (0)	100.00 a ± 0 (0)	5.07 a ± 0.04 (0)	2.90 a ± 0.02 (0)
	6.25	84.00 b ± 1 (16)	3.46 b ± 0.02 (32.29)	1.14 b ± 0.24 (61.35)	98.00 a ± 1.22 (2)	3.62 b ± 0.02 (27.6)	1.25 b ± 0.02 (57.48)	100.00 a ± 0 (0)	4.01 b ± 0.02 (20.91)	1.40 b ± 0.01 (51.72)
	12.5	39.00 c ± 1 (61)	1.77 c ± 0.33 (65.36)	0.69 c ± 0.01 (76.61)	74.00 b ± 1.87 (26)	2.01 c ± 0.34 (59.8)	0.91 c ± 0.04 (69.05)	82.00 b ± 2 (18)	2.85 c ± 0.23 (43.79)	1.30 c ± 0.12 (55.17)
	25	17.00 d ± 2.55 (83)	0.88 d ± 0.56 (82.78)	0.36 d ± 0.04 (87.80)	52.00 c ± 1.47 (48)	0.79 d ± 0.07 (84.2)	0.63 d ± 0.02 (78.57)	55.00 c ± 1.58 (45)	2.03 d ± 0.71 (59.96)	1.09 d ± 0.01 (62.41)
	50	4.00 e ± 1 (96)	0.19 e ± 0.49 (96.28)	0.12 e ± 0.03 (95.93)	20.00 d ± 1.58 (80)	0.50 e ± 0.06 (90)	0.31 e ± 0.01 (89.46)	25.00 d ± 2.74 (75)	1.08 e ± 0.58 (78.70)	0.41 e ± 0.57 (85.86)
	100	0.00 e ± 0 (100)	0.00 f ± 0 (100)	0.00 f ± 0 (100)	0.00 e ± 0 (100)	0.00 f ± 0.01 (100)	0.00 f ± 0 (100)	6.00 e ± 1 (94)	0.34 f ± 1.46 (93.29)	0.20 f ± 0.02 (93.10)

Data are expressed as mean ± standard error. Means with the same letters in the column for each extract are not significantly different at $p > 0.05$. Values inside the parenthesis are inhibition percentages relative to the control.

No survival rate was recorded in *A. conyzoides* and *E. hirta* when *P. hysterophorus* extract was applied at 50 g L^{-1}. Meanwhile, no significant inhibition on the survival rate of all weed species was observed when *C. rutidosperma* and *B. alata* extracts were applied at a low concentration of 6.25 g L^{-1}. However, *C. rutidosperma* and *B. alata* inhibited seed survival rate at the highest concentrations by 100% and 90%, respectively. The survival rate of *A. conyzoides* and *E. hirta* seeds was more sensitive to the extracts compared to Weedy rice and *E. colona* seeds.

The radicle length of the target weed was significantly reduced ($p < 0.05$) by *P. hysterophorus* extract at a concentration equal to or higher than 6.25 g L^{-1}. The root growth of *A. conyzoides*, *E. hirta*, *E. colona*, and Weedy rice was reduced by 89.3%, 85.4%, 89.5%, and 88.7% when treated with *C. rutidosperma* extracts at the concentration of 50 g L^{-1}. No radicle development of the test species was observed when *P. hysterophorus* extract was applied at the highest concentration, whereas up to 90% inhibition was observed by *B. alata* extract (Table 1). All extracts decreased the hypocotyl elongation of the target weeds. At the concentration of 50 g L^{-1}, *P. hysterophorus*, *C. rutidosperma*, and *B. alata* extracts reduced the hypocotyl length of all tested weeds by 95–100%, 84–90%, and 76–81%, respectively. Therefore, the extent of inhibition of *P. hysterophorus* extract was higher compared to *C. rutidosperma* and *B. alata* extracts.

3.1.2. Effect of Methanol Extracts on the Survival Rate and Initial Growth of Crops

The survival rate, hypocotyl and radicle length of the tested crops were also significantly influenced by the methanol extract of *P. hysterophorus*, *C. rutidosperma*, and *B. alata*. The decrement of these parameters increased with the increase of the extract concentration when compared to the control (Table 2).

The survival rate of *Z. mays*, *O. sativa*, *A. esculentus*, and *A. gangeticus* were reduced by 46%, 100%, 85%, and 100%, respectively for *P. hysterophorus* extracts, 35%, 93%, 64%, and 100%, respectively for *C. rutidosperma* extract, and 21%, 85%, 46%, and 88%, respectively for *B. alata* extract at the highest concentration i.e., 100 g L^{-1} (Table 2). *Parthenium hysterophorus* showed the highest phytotoxic effect on *A. gangeticus* (100%) compared to the other crops at 50 g L^{-1}.

The reduction in radicle length ranged from 93 to 100% for *P. hysterophorus* extract, 78 to 100% for *C. rutidosperma* extract, and 74 to 88% for *B. alata* extract at the highest concentration (100 g L^{-1}). Extracts of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* differed from each other in reducing the radicle length of tested crops compared to the control (Table 2). The *P. hysterophorus* extract exerted a higher effect in reducing the radicle length of the target crops. For instance, at 50 g L^{-1} of *P. hysterophorus* extract, the radicle growth of *A. gangeticus* was completely suppressed (100%) while in *C. rutidosperma* and *B. alata* extracts it was reduced by 89.2% and 77.4%, respectively.

Hypocotyl growth of all tested crops responded differently to *P. hysterophorus*, *C. rutidosperma*, and *B. alata* extracts. The highest concentration (100 g L^{-1}) of *P. hysterophorus* extract resulted in a reduction of 80 to 100% in hypocotyl length of the tested species. On the other hand, 100 g L^{-1} of *C. rutidosperma* and *B. alata* extracts resulted in 67 to 100% and 65 to 82% hypocotyl length reduction, respectively. At the lowest concentration (6.25 g L^{-1}), *C. rutidosperma* and *B. alata* did not show any significant effect on the hypocotyl growth of *Z. mays*. The hypocotyl length of test crops was reduced by arange of 10.8 to 100%, 1.4 to 100%, and 1.4 to 82.0% when treated with *P. hysterophorus*, *C. rutidosperma* and *B. alata* extracts, respectively.

Table 2. Effects of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* methanol extracts on survival rate (%), hypocotyl and radicle length (cm) of tested crops.

Tested Plants	Dose (g L^{-1})	*P. hysterophorus*			*C. rutidosperma*			*B. alata*		
		Survival Rate (%)	Hypocotyl Length (cm)	Radicle Length (cm)	Survival Rate (%)	Hypocotyl Length (cm)	Radicle Length (cm)	Survival Rate (%)	Hypocotyl Length (cm)	Radicle Length (cm)
O. sativa	0.00	100.00 a ± 0 (0)	3.16 a ± 0.02 (0)	6.17 a ± 0.16 (0)	100.00 a ± 0 (0)	3.19 a ± 0.03 (0)	6.17 a ± 0.01 (0)	100.00 a ± 0 (0)	3.18 a ± 0.03 (0)	6.19 a ± 0.24 (0)
	6.25	91.00 b ± 2.91 (9)	2.06 b ± 0.28 (34.81)	2.88 b ± 0.03 (53.32)	100.00 a ± 0 (0)	2.92 b ± 0.02 (8.46)	3.11 b ± 0.04 (49.59)	100.00 a ± 0 (0)	3.01 b ± 0.75 (5.35)	3.34 b ± 0.1 (46.04)
	12.5	58.00 c ± 3 (42)	1.03 c ± 0.22 (67.40)	2.00 c ± 0.02 (67.58)	86.00 b ± 1.87 (14)	1.94 c ± 0.1 (39.18)	3.31 c ± 0.42 (62.56)	88.00 b ± 1.22 (12)	2.14 c ± 0.16 (32.70)	2.41 c ± 0.03 (61.07)
	25	25.00 d ± 2.24 (75)	0.63 d ± 0.6 (80.06)	0.99 d ± 0.3 (83.95)	48.00 c ± 2 (52)	0.99 d ± 0.04 (68.96)	1.11 d ± 0.03 (82.01)	60.00 c ± 1.58 (40)	1.13 d ± 0.12 (64.46)	1.95 d ± 0.02 (68.50)
	50	10.00 e ± 1.58 (90)	0.42 e ± 0.01 (86.71)	0.55 e ± 0.26 (91.08)	22.00 d ± 1.22 (78)	0.70 e ± 0.13 (78.06)	0.87 e ± 0.02 (85.89)	31.00 d ± 1.87 (69)	0.87 e ± 0.13 (72.64)	1.15 e ± 0.1 (81.42)
	100	0.00 f ± 0 (100)	0.00 f ± 0 (100)	0.00 f ± 0 (100)	7.00 e ± 1.24 (93)	0.44 f ± 0.19 (86.21)	0.24 f ± 0.02 (96.11)	15.00 e ± 1.58 (85)	0.68 f ± 0.71 (78.62)	0.88 f ± 0.33 (85.78)
Z. mays	0.00	100.00 a ± 0 (0)	2.88 a ± 0.05 (0)	5.98 a ± 0.02 (0)	100.00 a ± 0 (0)	2.79 a ± 0.3 (0)	5.90 a ± 0.23 (0)	100.00 a ± 0 (0)	2.83 a ± 0.25 (0)	5.89 a ± 0.24 (0)
	6.25	100.00 a ± 0 (0)	2.57 b ± 0.01 (10.76)	4.91 b ± 0.03 (17.89)	100.00 a ± 0 (0)	2.75 a ± 0.14 (1.43)	5.10 b ± 0.02 (13.56)	100.00 a ± 0 (0)	2.79 a ± 0.59 (1.41)	5.13 b ± 0.16 (12.90)
	12.5	92.00 ab ± 2 (8)	2.14 c ± 0.02 (25.69)	3.28 c ± 0.25 (45.15)	100.00 a ± 0 (0)	2.48 b ± 0.02 (11.11)	3.88 ± 0.22 (34.24)	100.00 a ± 0 (0)	2.31 b ± 0.21 (18.37)	3.91 c ± 0.35 (33.62)
	25	85.00 b ± 2.24 (15)	1.15 c ± 0.2 (60.07)	1.98 d ± 0.31 (66.89)	90.00 b ± 1.58 (10)	2.03 c ± 0.04 (27.24)	2.91 ± 0.35 (50.68)	95.00 a ± 1.58 (5)	2.15 c ± 0.13 (24.03)	2.38 d ± 0.01 (59.59)
	50	75.00 c ± 3.53 (25)	0.91 e ± 0.35 (68.40)	1.02 e ± 0.04 (82.94)	74.00 c ± 1.87 (26)	1.30 d ± 0.01 (53.41)	1.26 ± 0.01 (78.64)	87.00 b ± 2.55 (13)	1.16 d ± 0.15 (59.01)	1.56 e ± 0.02 (73.51)
	100	54.00 d ± 1.87 (46)	0.56 f ± 0.31 (80.56)	0.39 f ± 0.47 (93.48)	65.00 d ± 3.53 (35)	0.91 e ± 0.04 (67.38)	0.77 ± 0.04 (86.95)	79.00 c ± 1.87 (21)	0.99 e ± 0.43 (65.02)	0.71 f ± 0.03 (87.95)

Table 2. *Cont.*

Tested Plants	Dose (g L^{-1})	*P. hysterophorus*			*C. rutidosperma*			*B. alata*		
		Survival Rate (%)	Hypocotyl Length (cm)	Radicle Length (cm)	Survival Rate (%)	Hypocotyl Length (cm)	Radicle Length (cm)	Survival Rate (%)	Hypocotyl Length (cm)	Radicle Length (cm)
A. esculentus	0.00	100.00 a ± 0 (0)	8.71 a ± 0.04 (0)	5.00 a ± 0.03 (0)	100.00 a ± 0 (0)	8.78 a ± 0.11 (0)	5.09 a ± 0.02 (0)	100.00 a ± 0 (0)	8.62 a ± 0.1 (0)	5.02 a ± 0.33 (0)
	6.25	100.00 a ± 0 (0)	6.52 b ± 0.03 (25.14)	3.23 b ± 0.15 (35.40)	100.00 a ± 0 (0)	7.05 b ± 0.04 (19.70)	4.00 b ± 0.03 (21.41)	100.00 a ± 0 (0)	7.67 b ± 0.03 (11.02)	4.28 b ± 0.15 (14.74)
	12.5	91.00 b ± 1.87 (9)	4.44 c ± 0.01 (49.02)	2.64 c ± 0.35 (47.20)	100.00 a ± 0 (0)	5.66 c ± 0.18 (35.54)	3.14 c ± 0.01 (38.31)	100.00 a ± 0 (0)	6.77 c ± 0.02 (21.46)	3.98 c ± 0.03 (20.72)
	25	81.00 c ± 1.9 (19)	2.94 d ± 0.25 (66.24)	1.96 d ± 0.01 (60.80)	90.00 a ± 3.16 (10)	3.91 d ± 0.29 (55.47)	2.35 d ± 0.19 (53.83)	94.00 a ± 2.45 (6)	5.20 d ± 0.81 (39.67)	3.08 d ± 0.26 (38.64)
	50	50.00 d ± 1.58 (50)	1.85 e ± 0.03 (78.76)	1.05 e ± 0.1 (79.00)	70.00 b ± 5.47 (30)	2.84 e ± 0.02 (67.65)	1.98 e ± 0.29 (61.10)	78.00 b ± 3.74 (22)	4.24 e ± 0.16 (50.81)	2.15 e ± 0.13 (57.17)
	100	15.00 e ± 1.5 (85)	1.09 f ± 0.12 (87.48)	0.29 f ± 0.09 (94.20)	36.00 c ± 4 (64)	1.90 f ± 0.19 (78.34)	1.09 f ± 0.12 (78.58)	54.00 c ± 2.45 (46)	3.00 f ± 0.04 (65.20)	1.29 f ± 0.06 (74.30)
A. gangeticus	0.00	100.00 a ± 0 (0)	3.10 a ± 0.16 (0)	2.02 a ± 0.02 (0)	100.00 a ± 0 (0)	3.09 a ± 0.01 (0)	2.04 a ± 0.18 (0)	100.00 a ± 0 (0)	3.06 a ± 0.03 (0)	2.08 a ± 0.25 (0)
	6.25	86.00 b ± 1.87 (14)	2.33 b ± 0.23 (24.84)	1.20 b ± 0.21 (40.59)	100.00 a ± 0 (0)	2.85 b ± 0.16 (7.77)	1.50 b ± 0.24 (26.47)	100.00 a ± 0 (0)	3.02 a ± 0.28 (4)	1.86 b ± 0.18 (10.58)
	12.5	70.00 c ± 3.53 (30)	1.15 c ± 0.39 (62.90)	0.44 c ± 0.01 (78.22)	80.00 b ± 1.58 (20)	1.55 c ± 0.02 (49.84)	0.85 c ± 0.33 (58.33)	88.00 b ± 2.55 (12)	2.28 b ± 0.16 (25.49)	0.96 c ± 0.2 (53.85)
	25	30.00 d ± 1.58 (70)	0.81 d ± 0.05 (73.87)	0.18 d ± 0.09 (91.09)	44.00 c ± 2.45 (56)	1.05 d ± 0.21 (66.02)	0.51 d ± 0.24 (75.00)	53.00 c ± 1.22 (47)	1.67 c ± 0.02 (45.42)	0.76 d ± 0.01 (63.46)
	50	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	11.00 d ± 1 (89)	0.58 e ± 0.27 (81.23)	0.22 e ± 0.11 (89.21)	23.00 d ± 1.24 (77)	1.02 d ± 0.17 (66.67)	0.47 e ± 0.06 (77.40)
	100	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 e ± 0 (100)	0.00 f ± 0 (100)	0.00 f ± 0 (100)	12.00 e ± 1.47 (88)	0.55 e ± 0.31 (82.03)	0.23 f ± 0.23 (88.94)

Data are expressed as mean ± standard error. Means with the same letters in the column for each extract are not significantly different at $p > 0.05$. Values inside the parenthesis are inhibition percentages relative to the control.

3.1.3. Comparison of Methanol Extracts on Examined Initial Growth Parameters and Plants

The half inhibitory concentrations of each extract for all test species are shown in Table 3. The effectiveness of the *P. hysterophorus* extract was higher than the *C. rutidosperma* and *B. alata* extract, as the rank value of *C. rutidosperma* extract (598.3 g L^{-1}) and *B. alata* extract (876.9 g L^{-1}) were more than the *P. hysterophorus* (393.9 g L^{-1}). The obtained EC_{50} showed differences among the response of test plants to the inhibitory effect of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* (Table 3). The differences in the sensitivity of species to the extracts were also evident from the rank values of plants. *Zea mays* was the only species affected at higher concentrations i.e., 157.7, 206.3, and 329.3 g L^{-1} of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* extracts, respectively. In other words, the *Z. mays* plant showed more tolerance, which indicates that only a high concentration of extracts could suppress this plant. The second less sensitive test plant (after *Z. mays*) was *A. esculentus*. The rank value of *A. conyzoides*, *E. hirta*, *E. colona*, Weedy rice, *O. sativa*, and *A. gangeticus* was 22.0, 22.6, 25.7, 26.6, 30.9, and 33.8 g L^{-1}, respectively, when they were treated with *P. hysterophorus* extract. The rank values for these tested plant species were 39.9, 44.2, 39.9, 39.9, 54.3, and 50.9 g L^{-1}, respectively for *C. rutidosperma* and 50.6, 51.6, 53.6, 55.0, 66.9, and 79.8 g L^{-1}, respectively for *B. alata* extracts. Therefore, it was apparent that these tested plant species were most sensitive to *P. hysterophorus* extract.

Table 3. EC_{50} and rank value (Re) of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* methanol extract for the tested species.

Target Plants	*P. hysterophorus*				*C. rutidosperma*				*B. alata*			
	ECs50	ECh50	ECr50	Rank (Re)	ECs50	ECh50	ECr50	Rank (Re)	ECs50	ECh50	ECr50	Rank (Re)
	g L^{-1}											
A. conyzoides	10.93	7.15	3.96	22.04	22.39	11.28	6.27	39.94	26.59	17.26	6.77	50.62
E. hirta	11.14	6.91	4.59	22.64	23.48	11.01	9.73	44.22	27.81	16.44	7.38	51.63
E. colona	11.73	9.38	4.54	25.65	23.84	10.59	5.42	39.85	28.60	16.89	8.08	53.57
Weedy rice	13.47	8.72	4.43	26.62	21.88	11.67	6.38	39.93	27.55	17.90	9.59	55.04
O. sativa	15.71	9.01	6.13	30.85	27.84	19.85	6.63	54.32	34.83	24.73	7.33	66.89
A. gangeticus	15.65	10.73	7.38	33.76	22.59	16.83	11.46	50.88	30.64	31.71	17.47	79.82
A. esculentus	46.77	14.72	13.21	74.7	74.56	23.47	24.83	122.86	104.70	47.32	37.98	190.00
Z. mays	115.46	26.24	15.96	157.66	131.86	52.07	22.34	206.27	254.07	50.44	21.77	329.28
Rank (Re)	240.86	92.86	60.24	393.92	348.44	156.77	93.06	598.27	534.79	222.69	116.37	876.85

ECs50, ECh50, and ECr50 are the concentration of extracts that inhibits 50% of germination, hypocotyl and radicle respectively and Rank is the sum of ECs50, ECh50, and ECr50.

3.2. Glasshouse Experiment

3.2.1. Effect of Methanol Extract on Plant Height, Root Length, Leaf Area, and Total Chlorophyll Content of Weeds

Data regarding the effect of the foliar spray of methanol extracts of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* on control (%), plant height, and root length of tested weeds are presented in Table 4. Among all the tested weeds, only *A. conyzoides* was controlled by 80% and 100% when sprayed with *P. hysterophorus* at a concentration of 50 g L^{-1} and 100 g L^{-1}, respectively. A similar trend was observed where an increase in the concentration of each treatment resulted in a remarkable reduction in plant height. Among the treatments, *P. hysterophorus* showed a more phytotoxic effect on the plant height of tested weeds compared to *C. rutidosperma* and *B. alata* at the highest concentration (100 g L^{-1}). At the same concentration, *P. hysterophorus* extract caused 100%, 60.0%, 20.4%, and 19.2% reduction in plant height of *A. conyzoides*, *E. hirta*, Weedy rice, and *E. colona*, respectively. On the other hand, 8.1to 11.1% and 5.6 to 8.6% plant height reduction was achieved by *C. rutidosperma* and *B. alata* extracts, respectively for all tested weeds.

Table 4. Effect of methanol extract of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* on the control (%), plant height (cm) and root length (cm) of *A. conyzoides*, *E. hirta*, weedy rice, and *E. colona*.

Tested Weeds	Dose (g L^{-1})	Control (%)			Plant Height (cm)			Root Length (cm)		
		PH	CR	BA	PH	CR	BA	PH	CR	BA
A. conyzoides	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	56.79 a ± 0.83 (0)	57.34 a ± 1.33 (0)	57.57 a ± 0.63 (0)	28.18 a ± 1.53 (0)	27.59 a ± 2.18 (0)	28.10 a ± 0.21 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	51.98 b ± 0.57 (8.45)	56.62 a ± 3.2 (1.27)	57.05 a ± 0.56 (0.90)	23.92 c ± 1.16 (15.09)	26.88 b ± 1.17 (2.56)	27.78 a ± 0.76 (1.12)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	46.11 b ± 0.79 (18.75)	56.40 a ± 3.4 (1.65)	57.02 a ± 2.37 (0.94)	21.38 c ± 0.19 (24.13)	26.77 b ± 2.21 (2.97)	27.70 a ± 2.51 (1.41)
	25	10.00 a ± 5.77	0.00 a ± 0	0.00 a ± 0	37.08 b ± 1.00 (34.72)	55.87 a ± 4.6 (2.58)	56.34 a ± 4.76 (2.13)	15.53 b ± 0.99 (44.91)	26.23 a ± 1.77 (4.92)	27.16 a ± 1.95 (3.36)
	50	80.00 a ± 8.16	0.00 b ± 0	0.00 b ± 0	13.30 b ± 4.46 (76.69)	54.03 a ± 0.49 (5.76)	55.45 a ± 0.78 (3.70)	4.65 b ± 1.57 (83.53)	25.10 a ± 0.89 (9.00)	26.23 a ± 0.67 (6.66)
	100	100.00 a ± 0	0.00 b ± 0	0.00 b ± 0	0.00 c ± 0 (100)	51.02 b ± 1.11 (11.03)	53.75 a ± 0.49 (6.63)	0.00 c ± 0 (100)	21.41 b ± 0.28 (22.40)	24.00 a ± 1.5 (14.56)
E. hirta	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	42.24 a ± 0.22 (0)	42.51 a ± 0.41 (0)	42.35 a ± 0.21 (0)	29.87 a ± 0.29 (0)	30.48 a ± 0.20 (0)	30.24 a ± 0.3 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	38.45 b ± 0.59 (8.98)	41.65 a ± 0.39 (2.01)	42.00 a ± 1.37 (0.82)	26.23 b ± 0.9 (12.18)	29.94 a ± 0.32 (1.77)	29.89 a ± 0.28 (1.13)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	35.93 b ± 3.45 (14.93)	41.52 a ± 0.38 (2.33)	41.74 a ± 0.78 (1.43)	24.16 b ± 0.11 (19.09)	29.37 a ± 1.31 (3.64)	29.63 a ± 0.39 (2.01)
	25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	31.01 b ± 4.63 (26.59)	41.43 a ± 0.45 (2.54)	41.39 a ± 1.21 (2.24)	20.23 b ± 0.58 (32.26)	28.76 a ± 0.28 (5.62)	28.98 a ± 0.32 (4.15)
	50	25.00 a ± 0	0.00 b ± 0	0.00 b ± 0	26.34 b ± 0.98 (37.64)	40.25 a ± 0.32 (5.31)	40.55 a ± 2.13 (4.23)	14.46 b ± 2.5 (51.61)	27.81 a ± 0.32 (8.76)	27.80 a ± 1.12 (8.07)
	100	55.00 a ± 0	0.00 b ± 0	0.00 b ± 0	16.51 b ± 0.48 (60.91)	38.34 a ± 0.24 (9.79)	38.71 a ± 0.69 (8.59)	8.79 b ± 0.65 (70.58)	24.50 a ± 0.88 (19.62)	25.39 a ± 0.45 (16.02)

Table 4. *Cont.*

Tested Weeds	Dose (g L^{-1})	Control (%)			Plant Height (cm)			Root Length (cm)		
		PH	CR	BA	PH	CR	BA	PH	CR	BA
Weedy rice	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	79.64 a ± 2.56 (0)	80.19 a ± 0.72 (0)	80.04 a ± 2.12 (0)	27.01 a ± 1.88 (0)	26.79 a ± 0.64 (0)	26.66 a ± 0.24 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	77.86 b ± 1.93 (2.24)	79.55 a ± 2.89 (0.80)	79.92 a ± 0.54 (0.16)	25.57 b ± 0.37 (5.32)	26.25 a ± 0.85 (2.01)	26.32 a ± 0.03 (1.29)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	77.51 c ± 1.31 (2.67)	78.72 b ± 1.83 (1.84)	79.48 a ± 1.48 (0.70)	24.18 c ± 1.12 (10.47)	25.77 b ± 0.14 (3.80)	26.21 a ± 0.11 (1.69)
	25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	73.61 c ± 0.67 (7.58)	77.74 b ± 0.65 (3.06)	78.65 a ± 1.08 (1.74)	22.54 b ± 0.23 (16.55)	25.32 a ± 1.2 (5.50)	25.44 a ± 1.2 (4.58)
	50	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	69.54 b ± 1.81 (12.68)	76.29 a ± 2.66 (4.87)	77.02 a ± 0.75 (3.78)	20.09 b ± 0.43 (25.60)	23.93 a ± 0.77 (10.68)	24.61 a ± 0.25 (7.69)
	100	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	63.38 c ± 1.33 (20.41)	73.72 b ± 1.87 (8.07)	75.55 a ± 0.98 (5.61)	15.66 b ± 0.26 (41.99)	21.96 a ± 0.42 (18.03)	22.13 a ± 0.45 (17.02)
E. colona	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	81.77 a ± 0.25 (0)	82.05 a ± 0.72 (0)	82.47 a ± 1.23 (0)	20.22 a ± 0.03 (0)	20.11 a ± 0.11 (0)	19.99 a ± 0.21 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	80.61 b ± 1.09 (1.41)	81.62 ab ± 0.28 (0.53)	82.35 a ± 0.54 (0.14)	19.08 b ± 0.06 (5.61)	19.59 a ± 0.62 (2.56)	19.65 a ± 0.28 (1.68)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	79.06 b ± 1.13 (3.31)	81.12 a ± 1.83 (1.14)	82.03 a ± 1.47 (0.53)	18.20 b ± 0.27 (9.78)	19.50 a ± 0.41 (3.03)	19.57 a ± 0.03 (2.09)
	25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	75.52 b ± 0.62 (7.64)	80.12 a ± 0.55 (2.36)	81.50 a ± 0.8 (1.17)	16.35 b ± 0.13 (19.11)	18.93 a ± 0.68 (5.86)	18.88 a ± 0.67 (5.52)
	50	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	71.52 b ± 2.18 (12.53)	78.15 a ± 0.65 (4.77)	79.64 a ± 0.32 (3.43)	15.06 b ± 0.56 (25.52)	18.16 a ± 0.28 (9.69)	18.06 a ± 0.50 (9.62)
	100	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	66.10 b ± 0.79 (19.17)	75.38 a ± 0.87 (8.13)	76.89 a ± 0.21 (6.76)	13.07 b ± 0.21 (35.32)	16.90 a ± 0.63 (15.93)	17.07 a ± 0.76 (14.62)

Data are expressed as mean. Means ± standard error with same letters in the row for each extract are not significantly different at $p > 0.05$. Values inside the parenthesis are inhibition percentages relative to the control. Note: PH = *P. hysterophorus*, CR = *C. rutidosperma*, BA = *B. alata*.

Among the tested weeds, the root length of *A. conyzoides* was inhibited completely (100%) when sprayed with 100 g L^{-1} *P. hysterophorus* extract. The inhibition of root length of all tested weeds ranged from 35.3 to 100%, 15.9 to 22.4%, and 14.6 to 16.5% at the same concentration (100 g L^{-1}) of *P. hysterophorus*, *C. rutidosperma*, and *B. alata*, respectively.

Declined leaf area and total chlorophyll content of all tested weeds werealso observed with an increase in the foliar spray of methanol extracts of *P. hysterophorus*, *C. rutidosperma*, and *B. alata*. Similar to plant height and root length, the leaf area and total chlorophyll of *A. conyzoides* were most affected by the foliar spray of *P. hysterophorus* extract compared to the others (Table 5). Leaf area inhibition of *A. conyzoides*, *E. hirta*, Weedy rice, and *E. colona* ranged from 15 to 100%, 12 to 70%, 5.3 to 42.0%, and 5.6 to 35.3%, respectively when sprayed with an increased amount of *P. hysterophorus* extract. A similar trend was also observed for total chlorophyll. The inhibition percentage for leaf area and total chlorophyll of all tested weeds ranged from 15.3 to 19.4 and 12.0 to 18.90 when sprayed with the highest concentration of *C. rutidosperma* and *B. alata* extracts, respectively.

3.2.2. Effect of Methanol Extract on Total Fresh and Dry Weight of Weeds

Total fresh and dry weights of all tested weeds were significantly influenced by the foliar spray of *P. hysterophorus* extract in a concentration-dependent pattern compared to *C. rutidosperma* and *B. alata* extract (Table 5). The control obtained the highest fresh and dry weight. However, the reduction differed among the targeted species and the treatments. *Parthenium hysterophorus* extract reduced the fresh and dry weight of tested weeds from 35.3 to 100% and 43.0 to 100% at 100 g L^{-1} compared to the control, respectively. At 50 g L^{-1} concentration, the foliar spray of *P. hysterophorus* extract reduced 52.8% and 87.1% total fresh weight and 56.0% and 90% total dry weight of *E. hirta* and *A. conyzoides*, respectively. On the other hand, among the treatments, the highest fresh and dry weight was recorded when different concentrations of *C. rutidosperma* and *B. alata* extracts were applied on the tested weeds (Table 5).

3.2.3. Phytotoxic Effect of Methanol Extracts on Plant Height and Root Length of Crops

The effect of treatments on the development of tested crops at the maturity stage is shown in Table 6. The result indicated that the suppressive magnitude of applied extracts was species-dependent. Plant height of all tested crops except *Z. mays* was significantly influenced by the extract of *P. hysterophorus* compared to *C. rutidosperma* and *B. alata* extract. There was no significant difference between the activities of *C. rutidosperma* and *B. alata* at the lowest concentration. The highest plant height reduction (62.2%) occurred at 100 g L^{-1} concentration of *P. hysterophorus* for *A. gangeticus*, and only 20.4% plant height of *O. sativa* was reduced at the same concentration (Table 6).

Table 5. Effect of methanol extract of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* on leaf area (cm^2), total chlorophyll (SPAD), total fresh weight (g pot^{-1}), and total dry weight (g pot^{-1}) of *A. conyzoides*, *E. hirta*, weedy rice, and *E. colona*.

Tested Weeds	Dose (g L^{-1})	Leaf Area (cm^2)			Total Chlorophyll (SPAD)			Total Fresh Weight (g pot^{-1})			Total Dry Weight (g pot^{-1})		
		PH	**CR**	**BA**	**PH**	**CR**	**BA**	**PH**	**CR**	**BA**	**PH**	**CR**	**BA**
A. conyzoides	0	811.72 a ± 4.74 (0)	816.68 a ± 2.53 (0)	813.86 a ± 5.12 (0)	33.61 a ± 0.11 (0)	34.01 a ± 0.28 (0)	33.45 a ± 0.03 (0)	87.87 a ± 0.57 (0)	89.01 a ± 0.41 (0)	88.71 a ± 0.15 (0)	12.06 a ± 0.22 (0)	12.27 a ± 0.21 (0)	12.10 a ± 0.19 (0)
	6.25	682.81 b ± 1.85 (15.87)	811.44 a ± 2.22 (0.64)	813.04 a ± 5.10 (0.10)	29.87 a ± 0.02 (11.13)	33.14 a ± 0.03 (2.56)	33.10 a ± 0.02 (1.05)	76.18 b ± 0.44 (13.30)	87.25 a ± 0.56 (1.98)	87.69 a ± 0.31 (1.15)	10.66 b ± 0.23 (11.63)	11.94 a ± 0.25 (2.69)	12.02 a ± 0.20 (0.69)
	12.5	525.31 b ± 5.23 (35.28)	806.17 a ± 2.39 (1.29)	806.13 a ± 3.67 (0.95)	26.74 b ± 0.20 (20.45)	32.26 a ± 0.17 (5.16)	32.69 a ± 0.09 (2.28)	66.84 b ± 0.57 (23.92)	86.84 a ± 0.45 (2.44)	87.25 a ± 0.25 (1.65)	9.53 b ± 0.20 (20.95)	11.65 a ± 0.23 (5.07)	11.85 a ± 0.18 (2.09)
	25	319.16 b ± 6.92 (60.69)	785.42 a ± 3.01 (3.83)	791.36 a ± 3.58 (2.76)	24.43 c ± 0.05 (27.31)	30.16 b ± 0.01 (11.32)	31.44 a ± 0.15 (6.02)	48.62 b ± 0.80 (44.69)	84.03 a ± 0.48 (5.60)	84.81 a ± 0.44 (4.40)	7.16 b ± 0.18 (40.60)	11.23 a ± 0.19 (8.50)	11.49 a ± 0.21 (5.07)
	50	125.20 b ± 4.20 (84.54)	750.16 a ± 3.60 (8.15)	758.11 a ± 6.05 (6.85)	15.09 b ± 1.12 (54.96)	28.94 a ± 0.02 (14.91)	29.33 a ± 0.01 (12.31)	11.34 b ± 1.81 (87.05)	80.83 a ± 0.54 (9.91)	82.25 a ± 0.42 (7.28)	1.19 b ± 0.43 (89.97)	10.71 a ± 0.25 (12.70)	10.94 a ± 0.25 (9.62)
	100	0.00 c ± 0 (100)	675.67 b ± 3.06 (17.27)	716.05 a ± 1.90 (12.01)	0.00 b ± 0 (100)	26.68 a ± 0.03 (21.55)	26.65 a ± 0.02 (20.32)	0.00 b ± 0 (100)	74.08 a ± 0.24 (16.77)	75.96 a ± 0.84 (14.38)	0.00 b ± 0 (100)	9.89 a ± 0.20 (19.42)	9.98 c ± 0.19 (17.45)
E. hirta	0	116.96 a ± 0.27 (0)	117.25 a ± 0.11 (0)	117.02 a ± 0.69 (0)	44.45 a ± 0.30 (0)	45.07 a ± 0.20 (0)	44.79 a ± 0.24 (0)	23.37 a ± 0.16 (0)	24.05 a ± 0.25 (0)	23.76 a ± 0.22 (0)	7.70 a ± 0.06 (0)	7.88 a ± 0.08 (0)	7.69 a ± 0.43 (0)
	6.25	100.24 b ± 0.40 (14.29)	116.33 a ± 0.14 (0.78)	116.82 a ± 0.12 (0.17)	40.43 b ± 0.16 (9.03)	44.33 a ± 0.07 (1.64)	44.49 a ± 0.23 (0.66)	20.80 b ± 0.13 (10.99)	23.36 a ± 0.27 (2.87)	23.35 a ± 0.47 (1.73)	6.73 b ± 0.04 (12.63)	7.50 a ± 0.05 (4.75)	7.51 a ± 0.45 (2.40)
	12.5	85.91 b ± 1.05 (26.54)	115.54 a ± 0.26 (1.45)	115.59 a ± 0.26 (1.22)	37.60 b ± 0.18 (15.41)	43.85 a ± 0.11 (2.71)	43.75 a ± 0.25 (2.33)	18.41 b ± 0.11 (21.21)	23.14 a ± 0.26 (3.81)	23.32 a ± 0.27 (1.86)	5.93 b ± 0.03 (23.01)	7.39 b ± 0.04 (6.15)	7.39 b ± 0.62 (3.90)
	25	71.46 b ± 0.49 (38.90)	112.79 a ± 0.63 (3.80)	112.94 a ± 0.23 (3.49)	35.30 b ± 0.01 (20.59)	43.00 a ± 0.04 (4.59)	42.52 a ± 0.21 (5.07)	13.62 b ± 1.08 (41.68)	22.52 a ± 0.28 (6.39)	23.02 a ± 0.17 (3.13)	4.50 b ± 0.13 (41.53)	7.19 a ± 0.21 (8.72)	7.28 a ± 0.41 (5.33)
	50	58.58 b ± 0.27 (49.91)	108.54 a ± 0.86 (7.43)	109.56 a ± 0.38 (6.37)	30.30 c ± 0.08 (31.83)	41.19 a ± 0.17 (8.61)	40.37 b ± 0.27 (9.87)	11.04 b ± 0.10 (52.75)	21.99 a ± 0.29 (8.58)	22.23 a ± 0.24 (6.43)	3.39 b ± 0.12 (55.97)	6.93 a ± 0.34 (12.08)	6.99 a ± 0.18 (9.07)
	100	38.95 b ± 0.96 (66.70)	99.28 a ± 0.72 (15.32)	100.44 a ± 1.29 (14.17)	21.88 b ± 0.58 (50.75)	36.77 a ± 0.21 (18.41)	37.37 a ± 0.24 (16.57)	7.04 b ± 0.11 (69.88)	20.37 a ± 0.30 (15.31)	20.41 a ± 0.19 (14.08)	1.92 b ± 0.06 (75.13)	6.43 a ± 0.64 (18.42)	6.41 a ± 0.49 (16.64)
Weedy rice	0	227.23 a ± 0.19 (0)	226.87 a ± 0.13 (0)	226.96 a ± 0.97 (0)	33.65 a ± 0.18 (0)	34.04 a ± 0.18 (0)	33.74 a ± 0.22 (0)	85.74 a ± 0.08 (0)	85.56 a ± 0.14 (0)	85.27 a ± 0.14 (0)	20.03 a ± 0.15 (0)	20.44 a ± 0.12 (0)	20.28 a ± 0.03 (0)
	6.25	220.65 b ± 0.30 (2.89)	225.79 a ± 0.23 (0.47)	225.81 a ± 0.24 (0.51)	32.03 b ± 0.54 (4.81)	33.41 a ± 0.12 (1.87)	33.52 a ± 0.64 (0.64)	79.14 b ± 0.17 (7.69)	83.95 a ± 0.07 (1.88)	84.17 b ± 0.12 (1.28)	18.85 b ± 0.08 (5.87)	19.98 a ± 0.01 (2.26)	20.06 a ± 0.49 (1.06)
	12.5	210.58 b ± 0.47 (7.33)	222.16 a ± 0.27 (2.08)	222.51 a ± 0.25 (1.96)	28.81 c ± 0.24 (14.40)	32.26 b ± 0.22 (5.25)	33.01 b ± 0.32 (2.16)	74.60 b ± 0.22 (13.00)	81.86 a ± 0.14 (4.32)	81.54 a ± 0.22 (4.38)	17.54 b ± 0.18 (12.41)	19.54 a ± 0.03 (4.40)	19.67 a ± 0.07 (3.01)
	25	182.94 b ± 3.12 (19.50)	216.98 a ± 2.11 (4.36)	215.72 a ± 0.96 (4.95)	25.72 c ± 0.76 (23.56)	31.61 b ± 0.09 (7.14)	32.15 a ± 0.13 (4.70)	68.56 b ± 0.52 (20.03)	78.62 a ± 0.37 (8.11)	79.56 a ± 0.14 (6.69)	15.30 c ± 0.05 (23.62)	18.62 b ± 0.04 (8.89)	19.05 a ± 0.11 (6.07)
	50	156.16 c ± 0.35 (31.28)	204.41 b ± 1.99 (9.90)	210.51 a ± 0.31 (7.25)	24.34 c ± 0.32 (27.66)	29.06 b ± 0.07 (14.62)	29.88 a ± 0.11 (11.44)	59.51 b ± 0.12 (30.59)	74.00 a ± 0.26 (13.51)	74.12 a ± 0.23 (13.07)	13.62 b ± 0.07 (31.99)	16.87 a ± 0.2 (17.47)	17.15 a ± 0.33 (15.42)
	100	125.33 c ± 0.12 (44.84)	182.76 b ± 0.48 (19.44)	187.56 a ± 0.60 (17.36)	22.12 c ± 0.43 (34.26)	27.21 b ± 0.08 (20.07)	28.32 a ± 0.60 (16.06)	50.51 b ± 0.23 (41.09)	67.88 a ± 0.46 (20.66)	68.60 a ± 0.09 (19.55)	11.61 c ± 0.08 (42.02)	14.14 b ± 0.06 (30.81)	14.71 a ± 0.55 (27.47)

Table 5. *Cont.*

Tested Weeds	Dose (g L^{-1})	Leaf Area (cm^2)			Total Chlorophyll (SPAD)			Total Fresh Weight (g pot^{-1})			Total Dry Weight (g pot^{-1})		
		PH	CR	BA	PH	CR	BA	PH	CR	BA	PH	CR	BA
E. colona	0	337.61 a ± 0.29 (0)	338.02 a ± 0.10 (0)	337.05 a ± 0.44 (0)	32.90 a ± 0.55 (0)	32.81 a ± 0.52 (0)	33.05 a ± 0.27 (0)	167.46 a ± 0.26 (0)	168.42 a ± 0.28 (0)	168.36 a ± 0.21 (0)	41.17 a ± 0.29 (0)	41.77 a ± 0.20 (0)	41.89 a ± 0.06 (0)
	6.25	317.54 b ± 1.13 (5.95)	329.60 a ± 0.73 (2.49)	332.89 a ± 0.96 (1.23)	31.32 a ± 0.42 (4.79)	32.06 a ± 0.50 (2.29)	32.71 a ± 0.28 (1.04)	160.79 b ± 0.41 (3.98)	167.00 a ± 0.24 (0.84)	167.17 a ± 0.26 (0.71)	38.13 b ± 0.11 (7.38)	40.97 a ± 0.21 (1.93)	41.41 a ± 0.05 (1.13)
	12.5	304.54 b ± 2.27 (9.80)	326.85 a ± 1.42 (3.30)	324.64 a ± 0.94 (3.68)	30.11 b ± 0.50 (8.49)	31.28 ab ± 0.43 (4.64)	32.20 a ± 0.22 (2.58)	155.66 b ± 0.61 (7.04)	164.69 a ± 0.28 (2.21)	164.59 a ± 0.35 (2.24)	35.83 b ± 0.13 (12.95)	40.21 a ± 0.19 (3.73)	40.74 a ± 0.08 (2.75)
	25	273.88 b ± 1.43 (18.88)	319.60 a ± 0.73 (5.45)	317.62 a ± 1.33 (5.77)	27.09 b ± 0.39 (17.57)	30.09 a ± 0.45 (8.16)	31.15 a ± 0.25 (5.76)	144.59 b ± 0.93 (13.65)	159.70 a ± 0.18 (5.18)	161.44 a ± 0.25 (4.11)	31.57 b ± 0.06 (23.31)	39.39 a ± 0.27 (5.71)	39.75 a ± 0.66 (5.11)
	50	244.88 b ± 1.60 (27.47)	302.10 a ± 1.35 (3.63)	305.37 a ± 2.12 (9.40)	25.57 b ± 0.08 (22.21)	28.93 a ± 0.38 (11.82)	28.96 a ± 0.42 (12.40)	132.62 b ± 0.44 (20.80)	152.36 a ± 0.58 (9.53)	153.52 a ± 0.41 (8.81)	28.04 b ± 0.38 (31.88)	37.56 a ± 0.22 (10.07)	37.98 a ± 0.19 (9.33)
	100	194.72 b ± 0.52 (42.32)	274.46 a ± 0.13 (18.80)	273.37 a ± 2.11 (18.90)	18.09 b ± 0.28 (44.99)	25.53 a ± 0.09 (22.11)	26.23 a ± 0.31 (20.61)	108.79 b ± 1.17 (35.03)	139.82 a ± 0.33 (16.98)	141.25 a ± 0.57 (16.10)	23.46 b ± 0.18 (42.99)	33.64 a ± 0.38 (19.46)	34.48 a ± 0.17 (17.69)

Data are expressed as mean ± standard error. Means with the same letters in the row for each extract are not significantly different at $p > 0.05$. Values inside the parenthesis are inhibition percentages relative to the control. In the table, PH = *P. hysterophorus*, CR = *C. rutidosperma*, BA = *B. alata*.

Table 6. Effect of methanol extract of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* on the control (%), plant height (cm), and root length (cm) of *O. sativa*, *Z. mays*, *A. esculentus*, and *A. gangeticus*.

Tested Crops	Dose (g L^{-1})	Control (%)			Plant Height (cm)			Root Length (cm)		
		PH	CR	BA	PH	CR	BA	PH	CR	BA
O. sativa	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	77.07 a ± 0.56 (0)	76.99 a ± 0.65 (0)	77.05 a ± 0.17 (0)	27.38 a ± 0.13 (0)	27.34 a ± 0.22 (0)	27.72 a ± 0.45 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	75.50 b ± 0.32 (2.04)	76.50 a ± 1.18 (0.64)	76.75 a ± 0.22 (0.38)	26.05 b ± 0.73 (4.84)	26.93 a ± 0.42 (1.50)	27.3 a ± 1.22 (1.52)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	72.92 c ± 1.27 (5.38)	75.91 b ± 2.11 (1.41)	76.45 a ± 1.21 (0.77)	24.96 b ± 0.11 (8.85)	26.39 a ± 0.20 (3.46)	27.01 a ± 0.32 (2.56)
	25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	69.99 c ± 0.64 (9.19)	74.92 b ± 0.75 (2.69)	75.69 a ± 0.63 (1.76)	23.10 b ± 0.01 (15.64)	25.74 a ± 0.98 (5.83)	26.41 a ± 0.53 (4.73)
	50	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	67.60 c ± 1.6 (12.21)	73.70 b ± 1.22 (4.27)	74.50 a ± 1.86 (3.31)	20.87 b ± 0.32 (23.81)	24.48 a ± 1.22 (10.45)	25.21 a ± 0.31 (9.03)
	100	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	61.34 b ± 0.70 (20.40)	70.66 a ± 0.93 (8.22)	72.07 a ± 0.13 (6.46)	16.81 c ± 0.16 (38.61)	22.22 b ± 0.30 (18.73)	23.28 a ± 0.82 (16.00)

Table 6. *Cont.*

Tested Crops	Dose (g L^{-1})	Control (%)			Plant Height (cm)			Root Length (cm)		
		PH	CR	BA	PH	CR	BA	PH	CR	BA
Z. mays	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	132.84 a ± 1.14 (0)	131.73 a ± 0.40 (0)	130.14 a ± 0.51 (0)	38.29 b ± 0.71 (0)	38.78 a ± 1.07 (0)	38.64 a ± 0.21 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	129.45 a ± 0.26 (2.53)	129.58 a ± 0.21 (1.63)	129.73 a ± 2.42 (0.32)	36.99 b ± 1.17 (3.39)	38.17 a ± 0.85 (1.57)	38.02 a ± 0.19 (1.62)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	126.13 b ± 0.37 (5.03)	129.30 a ± 1.23 (1.85)	129.04 a ± 0.68 (0.84)	35.79 b ± 0.24 (6.52)	37.65 a ± 1.18 (2.89)	37.64 a ± 2.60 (2.60)
	25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	121.02 c ± 1.16 (8.87)	128.41 a ± 1.58 (2.52)	127.56 b ± 0.21 (1.98)	32.37 b ± 0.54 (15.46)	36.99 a ± 0.54 (4.59)	36.84 a ± 1.65 (4.67)
	50	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	110.99 b ± 0.57 (16.42)	125.03 a ± 0.98 (5.08)	125.50 a ± 0.41 (3.56)	30.00 b ± 0.71 (21.66)	35.34 a ± 0.87 (8.85)	35.52 a ± 0.84 (8.09)
	100	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	97.39 b ± 0.49 (26.67)	119.77 a ± 0.54 (9.09)	120.71 a ± 0.33 (7.25)	23.41 b ± 0.45 (38.88)	32.41 a ± 0.11 (16.41)	32.67 a ± 0.11 (15.44)
A. esculentus	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	30.13 a ± 1.26 (0)	29.68 a ± 0.80 (0)	29.46 a ± 0.97 (0)	29.61 a ± 0.64 (0)	29.91 a ± 0.80 (0)	30.01 a ± 1.10 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	27.27 b ± 1.15 (9.43)	28.99 a ± 0.68 (2.32)	29.22 ± 1.28 (0.81)	26.13 b ± 1.15 (11.74)	29.01 a ± 0.68 (2.98)	29.12 a ± 0.28 (2.96)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	26.04 c ± 0.49 (13.55)	28.60 b ± 1.13 (3.63)	29.00 a ± 1.10 (1.56)	22.74 c ± 0.49 (23.21)	28.19 b ± 1.13 (5.73)	29.00 a ± 0.32 (3.35)
	25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	21.60 b ± 0.28 (28.31)	28.14 a ± 0.53 (5.17)	28.50 a ± 2.44 (3.28)	18.29 c ± 0.55 (38.24)	27.16 b ± 0.53 (9.19)	27.92 a ± 0.44 (6.95)
	50	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	17.94 c ± 1.03 (40.43)	27.07 b ± 0.91 (8.78)	27.98 a ± 1.05 (5.02)	16.29 b ± 1.03 (45.00)	25.70 a ± 1.91 (14.06)	26.12 a ± 0.48 (12.96)
	100	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	15.09 b ± 0.46 (49.91)	25.50 a ± 0.17 (14.07)	26.39 a ± 0.44 (10.44)	11.28 b ± 0.46 (61.91)	23.62 a ± 0.02 (21.00)	23.40 a ± 1.14 (22.03)

Table 6. *Cont.*

Tested Crops	Dose (g L^{-1})	Control (%)			Plant Height (cm)			Root Length (cm)		
		PH	CR	BA	PH	CR	BA	PH	CR	BA
A. gangeticus	0	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	66.78 a ± 1.40 (0)	67.02 a ± 1.12 (0)	66.61 a ± 0.58 (0)	29.57 a ± 1.44 (0)	29.34 a ± 0.64 (0)	29.19 a ± 0.69 (0)
	6.25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	61.64 b ± 2.41 (7.68)	65.83 a ± 0.63 (1.78)	65.53 a ± 0.87 (1.62)	25.04 b ± 0.74 (15.32)	28.57 a ± 1.16 (2.64)	28.63 a ± 0.81 (1.92)
	12.5	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	58.50 b ± 0.31 (12.39)	65.26 a ± 0.10 (2.63)	65.15 a ± 1.29 (2.20)	21.18 c ± 1.01 (28.37)	28.18 b ± 0.43 (3.95)	28.53 a ± 0.61 (2.26)
	25	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	50.16 b ± 1.27 (24.87)	64.24 a ± 0.23 (4.15)	63.98 a ± 0.41 (3.95)	18.24 c ± 0.64 (38.33)	27.33 b ± 0.61 (6.86)	27.88 a ± 1.84 (4.50)
	50	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	37.16 b ± 0.76 (44.34)	62.71 a ± 1.18 (6.43)	62.48 a ± 1.04 (6.20)	13.91 b ± 0.91 (52.96)	26.54 a ± 1.11 (9.55)	26.18 a ± 0.18 (10.33)
	100	0.00 a ± 0	0.00 a ± 0	0.00 a ± 0	25.22 b ± 0.64 (62.22)	59.41 a ± 0.92 (11.36)	59.21 a ± 0.91 (11.11)	8.23 b ± 1.23 (72.15)	23.91 a ± 0.24 (18.52)	24.54 a ± 0.33 (15.94)

Data are expressed as mean ± standard error.Means with the same letters in the row for each extract are not significantly different at $p > 0.05$. Values inside the parenthesis are inhibition percentages relative to the control. Note: PH = *P. hysterophorus*, CR = *C. rutidosperma*, BA = *B. alata*.

Root lengths of all tested plants were significantly decreased by all the applied extracts. Among the species, root length was more reduced in *A. gangeticus* at 100 g L^{-1} concentration of *P. hysterophorus* with an inhibition index of 72.2% followed by 61.9%, 38.9%, and 38.6% in *A. esculentus*, *Z. mays*, and *O. sativa*, respectively. This indicates that the effects caused by the *P. hysterophorus* extract on the plant height and root length were more prominent at higher concentrations across the species. The *C. rutidosperma* and *B. alata* extracts were less phytotoxic on the plant height and root length of the tested crops compared to *P. hysterophorus* extract. The extract of *C. rutidosperma* inhibited the plant height and root length of the tested crops by 8.2 to 14.1% and 9.2 to 21.0%, respectively.

3.2.4. Phytotoxic Effect of Methanol Extracts on Leaf Area, Total Chlorophyll, Fresh and Dry Weight of Crops

Foliar spray of *P. hysterophorus*, *C. rutidosperma*,and *B. alata* extract had a significant effect on leaf area and chlorophyll content of the test species (Table 7). The effects of *P. hysterophorus* extracts showed a decline from 6.3 to 61.0% at the lowest (6.25 g L^{-1}) to the highest (100 g L^{-1}) concentrations on the leaf area of *A. esculentus*, while 3.88 to 37.97% was recorded in *O. sativa.* The chlorophyll content of all tested crops except *O. sativa* was significantly affected by the foliar spray of *P. hysterophorus* extract at the concentration of 6.25 g L^{-1}. The test crop *A. esculentus* showed a 17.3% decrease in chlorophyll content compared to *O. sativa* when sprayed with *P. hysterophorus* extract at the highest concentration (100 g L^{-1}). Leaf area and chlorophyll content of *A. gangeticus* was inhibited by 22.1% and 18.3% by *C. rutidosperma* extract and 16.9% and 19.0% by *B. alata* extract, respectively at 100 g L^{-1} concentration.

Table 7. Effect of methanol extract of *P. hysterophorus*, *C. rutidosperma*,and *B. alata* on leaf area(cm^2), total chlorophyll (SPAD), total fresh and dry weight (g pot^{-1}) of *O. sativa*, *Z. mays*, *A. esculentus*, and *A. gangeticus*.

Tested Crops	Dose (g L^{-1})	Leaf Area (cm^2)			Total Chlorophyll (SPAD)			Total Fresh Weight (g pot^{-1})			Total Dry Weight (g pot^{-1})		
		PH	CR	BA	PH	CR	BA	PH	CR	BA	PH	CR	BA
O. sativa	0	321.46 a ± 4.17 (0)	324.19 a ± 2.80 (0)	323.94 a ± 3.80 (0)	36.93 a ± 0.27 (0)	37.52 a ± 0.17 (0)	37.13 a ± 0.05 (0)	96.03 a ± 0.25 (0)	95.54 a ± 0.27 (0)	95.13 a ± 0.24 (0)	17.17 a ± 0.17 (0)	17.46 a ± 0.13 (0)	17.09 a ± 0.07 (0)
	6.25	309.02 a ± 5.01 (3.88)	321.99 a ± 2.35 (0.67)	323.29 a ± 3.84 (0.20)	34.73 b ± 0.15 (5.95)	36.74 a ± 0.21 (2.07)	36.80 a ± 0.37 (0.89)	89.67 b ± 0.16 (6.62)	94.30 a ± 0.64 (1.29)	94.34 a ± 0.22 (0.83)	15.77 b ± 0.15 (8.11)	16.88 a ± 0.1 (3.32)	16.66 a ± 0.06 (2.50)
	12.5	297.94 b ± 4.60 (7.32)	317.49 a ± 4.34 (2.08)	319.23 a ± 3.60 (1.45)	32.66 b ± 0.20 (11.54)	35.79 a ± 0.27 (4.59)	36.03 a ± 0.07 (2.98)	84.23 b ± 0.67 (12.28)	91.88 a ± 0.31 (3.83)	92.67 a ± 0.28 (2.58)	14.67 b ± 0.05 (14.48)	16.10 a ± 0.03 (7.76)	16.18 a ± 0.14 (4.60)
	25	271.77 b ± 6.45 (15.46)	312.09 a ± 2.70 (3.73)	315.53 a ± 4.19 (2.60)	29.13 b ± 0.25 (21.12)	34.90 a ± 0.68 (6.98)	35.17 a ± 0.06 (5.28)	78.13 b ± 0.74 (18.64)	88.69 a ± 0.57 (7.16)	90.53 a ± 0.25 (4.83)	13.15 b ± 0.08 (23.35)	15.43 a ± 0.05 (11.57)	15.59 a ± 0.31 (8.77)
	50	239.77 b ± 4.37 (25.42)	299.27 a ± 3.97 (7.69)	299.84 a ± 4.39 (7.45)	26.75 b ± 0.36 (27.54)	32.37 a ± 0.64 (13.70)	32.80 a ± 0.18 (11.67)	70.44 c ± 0.32 (26.64)	84.44 b ± 0.18 (11.62)	85.82 a ± 0.32 (9.78)	11.94 c ± 0.13 (30.45)	14.18 b ± 0.41 (18.76)	14.63 a ± 0.40 (14.37)
	100	199.36 b ± 3.49 (37.97)	273.47 a ± 2.62 (15.63)	279.34 a ± 4.42 (13.78)	24.25 b ± 0.31 (34.33)	30.22 a ± 0.09 (19.44)	30.55 a ± 0.17 (17.72)	62.41 b ± 0.25 (35.01)	76.52 a ± 0.71 (19.90)	78.23 a ± 0.47 (17.76)	10.39 b ± 0.21 (39.44)	12.28 a ± 0.91 (29.65)	12.74 a ± 0.09 (25.45)
Z. mays	0	2502.50 a ± 0.12 (0)	2503.48 a ± 0.68 (0)	2502.06 a ± 0.40 (0)	39.43 a ± 0.23 (0)	39.00 a ± 0.17 (0)	38.84 a ± 0.08 (0)	181.12 a ± 0.39 (0)	181.53 a ± 0.30 (0)	181.33 a ± 0.09 (0)	22.11 a ± 0.14 (0)	21.91 a ± 0.15 (0)	22.00 a ± 0.20 (0)
	6.25	2401.58 b ± 0.50 (4.03)	2488.35 a ± 2.78 (0.60)	2485.81 a ± 2.87 (0.65)	36.51 b ± 0.20 (7.40)	38.06 a ± 0.73 (2.41)	37.97 a ± 0.02 (2.24)	169.67 b ± 0.51 (6.32)	178.77 a ± 0.21 (1.52)	179.02 a ± 0.44 (1.28)	20.79 b ± 0.03 (5.95)	21.5 a ± 0.13 (1.88)	21.84 a ± 0.16 (0.73)
	12.5	2309.58 b ± 4.79 (7.71)	2464.60 a ± 2.53 (1.55)	2471.31 a ± 4.37 (1.23)	34.39 b ± 0.03 (12.78)	37.06 a ± 0.07 (4.98)	36.92 a ± 0.23 (4.92)	163.95 b ± 0.61 (9.48)	176.02 a ± 0.39 (3.03)	177.72 a ± 0.52 (2.00)	19.44 c ± 0.06 (12.06)	21.00 b ± 0.07 (4.15)	21.30 a ± 0.03 (3.19)
	25	2083.63 b ± 0.20 (16.74)	2405.10 a ± 6.45 (3.93)	2408.29 a ± 4.39 (3.75)	31.97 c ± 0.04 (8.93)	35.83 a ± 0.04 (8.13)	35.24 b ± 0.02 (9.25)	152.29 c ± 0.14 (15.92)	172.00 b ± 0.41 (5.25)	173.67 a ± 0.48 (4.23)	17.58 c ± 0.01 (20.49)	20.53 b ± 0.06 (6.27)	21.09 a ± 0.02 (4.11)
	50	1813.28 c ± 0.81 (27.54)	2291.75 b ± 0.15 (8.46)	2324.72 a ± 5.14 (7.09)	28.31 c ± 0.64 (28.11)	34.44 a ± 0.25 (11.71)	32.38 b ± 0.03 (16.62)	135.89 c ± 0.04 (24.97)	161.34 b ± 0.29 (11.12)	165.34 a ± 0.42 (8.82)	15.46 c ± 0.07 (30.08)	19.30 b ± 0.11 (11.88)	20.a ± 0.07 (6.76)
	100	1602.44 c ± 2.37 (35.97)	2108.13 b ± 0.66 (15.79)	2128.98 a ± 0.64 (14.91)	24.69 c ± 0.21 (37.39)	30.25 b ± 0.9 (22.43)	30.86 a ± 0.30 (20.53)	123.37 b ± 0.19 (31.88)	147.41 a ± 0.37 (18.80)	149.17 a ± 0.72 (17.74)	14.06 c ± 0.20 (36.40)	17.50 b ± 0.12 (20.13)	19.53 a ± 0.10 (11.21)
A. esculentus	0	1130.05 a ± 0.05 (0)	1129.76 a ± 0.28 (0)	1131.29 a ± 0.93 (0)	52.97 a ± 0.23 (0)	53.17 a ± 0.06 (0)	53.45 a ± 0.24 (0)	103.31 a ± 0.44 (0)	103.68 a ± 0.03 (0)	104.11 a ± 0.34 (0)	13.46 a ± 0.22 (0)	13.67 a ± 0.19 (0)	13.38 a ± 0.04 (0)
	6.25	1059.36 b ± 0.29 (6.26)	1113.51 a ± 2.20 (1.44)	1117.54 b ± 2.70 (1.22)	46.65 b ± 0.02 (11.94)	52.45 a ± 0.16 (1.35)	52.11 b ± 0.43 (2.49)	95.53 b ± 0.61 (7.53)	102.16 a ± 0.13 (1.47)	102.85 a ± 0.17 (1.21)	12.11 b ± 0.03 (9.92)	13.22 a ± 0.08 (3.26)	13.05 a ± 0.08 (2.48)
	12.5	982.41 b ± 2.15 (13.06)	1103.51 a ± 2.54 (2.32)	1099.79 a ± 4.09 (2.78)	44.12 b ± 0.04 (16.71)	51.29 a ± 0.04 (3.53)	50.80 a ± 0.23 (4.95)	89.43 b ± 0.28 (13.43)	101.51 a ± 0.19 (2.10)	101.81 a ± 0.15 (2.21)	10.82 b ± 0.31 (19.53)	12.91 a ± 0.09 (6.82)	12.55 a ± 0.39 (6.22)
	25	878.41 c ± 0.01 (22.27)	1067.40 b ± 1.31 (5.52)	1077.30 a ± 0.10 (4.77)	41.00 c ± 0.43 (22.60)	47.65 b ± 0.18 (10.37)	48.10 a ± 0.64 (10.00)	76.26 b ± 0.49 (26.17)	99.61 a ± 0.06 (3.93)	99.17 a ± 0.22 (4.75)	8.87 c ± 0.05 (34.04)	12.57 a ± 0.05 (8.80)	12.30 b ± 0.03 (8.11)
	50	611.10 c ± 0.13 (45.92)	1007.45 b ± 4.67 (10.83)	1044.80 a ± 4.83 (7.65)	33.20 c ± 0.91 (37.32)	44.89 b ± 0.43 (15.58)	45.80 a ± 0.45 (14.31)	64.18 b ± 0.37 (37.87)	95.59 a ± 0.31 (7.81)	96.60 a ± 0.41 (7.21)	6.84 b ± 0.04 (49.16)	12.12 a ± 0.46 (11.30)	12.13 a ± 0.10 (9.36)
	100	441.26 c ± 0.23 (60.95)	917.77 b ± 0.16 (18.76)	956.01 a ± 0.33 (15.49)	25.61 b ± 0.56 (51.67)	42.97 a ± 0.87 (19.18)	43.32 a ± 0.85 (18.93)	43.04 b ± 0.32 (58.33)	87.41 a ± 0.30 (15.70)	88.33 a ± 0.70 (15.15)	4.74 b ± 0.44 (64.72)	11.20 a ± 0.1 (17.99)	11.39 a ± 0.09 (14.85)

Table 7. *Cont.*

Tested Crops	Dose (g L^{-1})	Leaf Area (cm^2)			Total Chlorophyll (SPAD)			Total Fresh Weight (g pot^{-1})			Total Dry Weight (g pot^{-1})		
		PH	CR	BA	PH	CR	BA	PH	CR	BA	PH	CR	BA
A. gangeticus	0	1354.77 a ± 0.47 (0)	1354.08 a ± 0.08 (0)	1353.84 a ± 0.27 (0)	48.39 a ± 0.10 (0)	47.94 a ± 0.26 (0)	48.20 a ± 0.01 (0)	98.90 a ± 0.37 (0)	98.21 a ± 0.06 (0)	98.22 a ± 0.24 (0)	13.45 a ± 0.05 (0)	13.04 b ± 0.08 (0)	13.17 b ± 0.8 (0)
	6.25	1209.63 b ± 0.79 (10.71)	1333.94 a ± 4.35 (1.49)	1331.78 a ± 0.64 (1.63)	41.86 b ± 0.05 (13.48)	47.51 a ± 0.24 (0.88)	47.24 a ± 0.18 (1.99)	87.27 b ± 0.32 (11.75)	96.30 a ± 0.19 (1.94)	96.31 a ± 0.12 (1.94)	11.67 b ± 0.06 (13.25)	12.77 a ± 0.07 (2.07)	12.75 a ± 0.05 (3.18)
	12.5	1111.88 c ± 2.14 (17.93)	1302.04 b ± 0.83 (3.84)	1313.19 a ± 4.14 (3.00)	39.18 b ± 0.04 (19.02)	46.76 a ± 0.19 (2.44)	46.75 a ± 0.14 (3.00)	75.47 b ± 0.04 (23.68)	94.46 a ± 0.36 (3.82)	95.08 a ± 0.52 (3.20)	10.26 b ± 0.6 (23.70)	12.64 a ± 0.18 (3.05)	12.61 a ± 0.22 (4.22)
	25	958.32 c ± 0.11 (29.26)	1254.79 b ± 4.80 (7.33)	1292.78 a ± 3.89 (4.51)	32.24 b ± 0.02 (33.37)	45.51 a ± 0.24 (5.06)	45.60 a ± 0.03 (5.38)	64.29 b ± 0.38 (34.99)	92.01 a ± 0.24 (6.32)	92.89 a ± 0.28 (5.43)	8.45 b ± 0.18 (37.19)	12.38 a ± 0.54 (5.08)	12.47 a ± 0.14 (5.25)
	50	807.70 c ± 3.20 (40.38)	1144.79 b ± 2.51 (15.45)	1242.28 a ± 4.09 (8.24)	28.49 b ± 0.49 (41.12)	43.01 a ± 0.40 (10.28)	43.35 a ± 0.25 (10.05)	50.19 c ± 0.07 (49.25)	89.05 b ± 0.26 (9.33)	90.32 a ± 0.44 (8.04)	6.94 b ± 0.1 (48.38)	12.08 a ± 0.11 (7.30)	11.87 a ± 0.11 (9.83)
	100	550.20 c ± 1.77 (59.39)	1054.91 b ± 1.65 (22.09)	1135.31 a ± 2.65 (16.14)	24.93 b ± 0.24 (48.47)	39.18 a ± 0.03 (18.26)	39.04 a ± 0.07 (19.01)	38.52 c ± 0.82 (61.04)	82.07 b ± 0.22 (16.43)	84.83 a ± 0.54 (13.64)	5.63 b ± 0.07 (58.14)	11.26 a ± 0.10 (13.67)	11.10 a ± 0.09 (15.66)

Data are expressed as mean ± standard error. Means with the same letters in the row for each extract are not significantly different at $p > 0.05$. Values inside the parenthesis are inhibition percentages relative to the control. Note: PH = *P. hysterophorus*, CR = *C. rutidosperma*, BA = *B. alata*.

The foliar spray of *P. hysterophorus*, *C. rutidosperma*, and *B. alata* also had a significant effect on the total fresh and dry weight of all tested species and the effect was concentration-dependent (Table 7). Moreover, the effect of extracts on different tested species at the same concentration was varied. Total dry weight was decreased from 9.9 to 64.7% in *A. esculentus* followed by 13.3 to 58.1%, 8.1 to 39.4%, and 6.0 to 36.4% in *A. gangeticus*, *O. sativa*, and *Z. mays*, respectively with a foliar spray of *P. hysterophorus* extract at lowest (6.25 g L^{-1}) to highest concentrations (100 g L^{-1}). For *A. gangeticus*, 61.1% growth reduction was achieved by *P. hysterophorus* extract while 16.4% and 13.6% reduction were achieved by *C. rutidosperma* and *B. alata* extract, respectively. On the other hand, *A. gangeticus* obtained a relatively higher reduction (61.0%) of total fresh weight for *P. hysterophorus* extract compared to other tested species and applied extracts.

4. Discussion

The methanol extract of three Malaysian invasive weeds had the ability to affect survival rate (%) and seedling growth of four selected weed species (*A. conyzoides*, *E. hirta*, Weedy rice, and *E. colona*) and four crops (*Z. mays*, *O. sativa*, *A. esculentus*, and *A. gangeticus*), under laboratory conditions. All these extracts influenced the survival rate, hypocotyl and radicle growth of tested species in a dose-dependent manner. Extracts of *P. hysterophorus* and *C. rutidosperma* were the most promising because of their remarkable strength, potency, and regularity in inhibiting germination and seedling growth of all tested species. The inhibition of plant extracts on the germination process is thought to be associated with osmotic effects on the rate of imbibition, which ultimately inhibits the initiation of germination and, especially, cell elongation [25]. The *P. hysterophorus* extract at 50 g L^{-1} fully inhibited the seed germination and seedling growth of *A. conyzoides*, *E. hirta*, and *A. gangeticus*. The inhibitory effect of *P. hysterophorus* extract and residues on the growth and development of some field crops were also reported by Batish et al. [27], Singh et al. [28], Mersie and Singh [29].

Moreover, at 50 g L^{-1} concentration, *C. rutidosperma* also caused significant inhibition of all tested seeds. Ladhari et al. [30] reported the allelopathic properties of *Cleome arabica* L. and identified 11-α-acetylbrachy-carpone-22(23)-ene as the main allelopathic compound. Whereas, Ahmed et al. [31] stated that the asdamarane type triterpene, for instance, 11-α-acetylbrachy-carpone-22(23)-ene, 17-α-hydroxycabraleactone, and amblyone were responsible for the toxicity of *Cleome amblyocarpa*.

The inhibitory effect of the test extracts varied among the eight species examined, and *A. conyzoides* was more sensitive to tested extracts than the other tested plants. The present study is in agreement with Ishak and Sahid [32] who found that the extract of *Leucaena leucocephala* at 66.7 g L^{-1} reduced germination, hypocotyl, and radicle elongation of *A. conyzoides* by 48%, 47%, and 65%, respectively. Furthermore, the radicle length of the tested species was more sensitive to extracts compared to the germination percentage and the hypocotyl length. The greater sensitivity of radicle growth to the allelopathic plant extracts is because—radicles are the first organ that are exposed to the phytotoxic substances and a more highly permeable tissue than other organs [19,33,34], and/or a low mitotic division in the root apical meristem [35]. Moreover, the allelopathic substances can affect genes responsible for the cellular characterization of radicle tissues and endoderm, reducing its development [36].

The glasshouse experiment provided further evidence for the higher allelopathic potential of *P. hysterophorus* extract compared to *C. rutidosperma* and *B. alata* as observed in the laboratory. The results showed that *P. hysterophorus* extract at 50 g L^{-1} and 100 g L^{-1} significantly reduced the growth of 21-days-old *A. conyzoides* and *E. hirta*. The highest reduction of 100% was observed from the maximum concentration (100 g L^{-1}) of *P. hysterophorus* extract at the mature stage of *A. conyzoides*. This type of dose-dependent inhibitory activity was reported by many researchers around the globe [19,34,37,38]. A greater decrease in plant height was recorded in *A. conyzoides* compared to other species. At 21 days after spray, only untreated *A. conyzoides* began flowering which indicates that the other treated plants might be suppressed by allelochemicals stress. Aslam et al. [39] reported the phytotoxic effect of *Calatropis procera*, *Peganum harmala*, and *Tamarix aphylla* on the shoot and root length of mustard and wheat, and wheat was sensitive to all three extracts at all the concentrations. Mulberry aqueous leaf extract suppressed shoot and root length, shoot and root dry matters of Bermuda grass by 90% and 80% at 100% concentrations, respectively [40]. Hassan et al. [41] also observed a decrease in shoot and root length of *Zea mays* and *Vigna unguiculata* treated with increased concentrations of *Jatropha curcas* extract.

Foliar spray of *P. hysterophorus* extract reduced dry weights and leaf area as the level of concentration increased across species although the species responded independently. The reduction in total dry weight was observed to be associated with a decrease in plant height and leaf area. Total dry weight and leaf area were mostly decreased in *A. conyzoides* and *E. hirta*, respectively. Leaf area reduction was higher in *A. conyzoides* and lower in *Z. mays* at 21 days after spraying with *P. hysterophorus* extract. This type of species-dependent inhibitory activity was also reported by several studies. For example, phytotoxins have an adverse impact on the growth of certain plants while having little or no inhibition in other plants at certain concentrations [42–44]. Several studies reveal a decline in leaf area of certain plant species using different extracts [45,46].

Chlorophyll is a determinant factor in photosynthesis and it was found to be lower in *A. conyzoides* among all tested species. The leaves of the tested plants appeared partially folded and this may lead to a decrease in photosynthetic activity [47]. Reduction of chlorophyll content in plants due to application of allelopathic plant extracts was also reported by Kamal [48], Siyar et al. [49], and Abdel-Farid [50].

It was also observed in the present study that the application of plant extracts in a foliar spray in laboratory conditions caused more inhibition compared to glasshouse conditions. Similar findings were also reported by Al-Humaid and El-Mergawi [21]. The inhibition by foliar spray may occur through various mechanisms such as suppressed hormone activity, a decreased rate of ion absorption, enzyme activity inhibition, reduce cell membrane permeability and also inhibit certain physiological processes such as photosynthesis, respiration, and protein formation. Thus, the seedling stage and the more mature stage of target plants vary in their sensitivities to plant extracts.

5. Conclusions

The study demonstrated that all the methanol extracts from three Malaysian invasive weeds (*P. hysterophorus*, *C. rutidosperma*, and *B. alata*) have allelopathic potential on the seed germination, growth, and development of tested plants. *P. hysterophorus* appeared as the most phytotoxic plant extract among the three. Moreover, the phytotoxic effect of the extracts was dependent on the target species, extract concentrations, and the extracted plant species. The growth and development of the tested plant species in the glasshouse were less affected compared to seed germination and growth under laboratory conditions. The only phytotoxic impact was provided by *P. hysterophorus* on the tested plant species in the glasshouse trial. Among the test species, *A. conyzoides* was more sensitive to *P. hysterophorus* extract. Taking into account the promising result of *P. hysterophorus* extract, this weed could be used for further study to develop a natural product-based herbicide for sustainable green agriculture. Identification and characterization of the most active phytotoxic compounds of the *P. hysterophorus* extract will be the first step of future studies.

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