

ORIGINAL ARTICLES

Allelopathic Effects of Croton Leaves Residue on Jasmine Trees and Associated Weeds

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ABSTRACT

Growth inhibitory effects of aqueous extracts of leaves and stems of *Croton (Codiameum variegatum pactum L.)* on *Portulaca oleracea* (Broadleaved weed) and *Echinochloa colonum* (grass) were investigated. Both extracts of croton caused inhibition of the seedling length of the two weeds. Growth inhibition was more pronounced in the presence of leaf extracts than the stem extract. Croton leaves residue was investigated in field studies 2008 and 2009 to find out its effect on weeds surrounding jasmine trees. Leaf residue with different rates from 50 up to 300 g/m² caused varying degrees of inhibition of broadleaved and grassy weeds 60 days after treatment and at 120 days, higher rates were more inhibition. The inhibition continued during the experimental period. On the other hand, the inhibition of weeds was accompanied with increase in flower yield as well as dry weight of 100 flowers. The results also revealed increase in concrete of flowers and percentage of oil yield. Estimation of micro and macronutrients resulted in increase in their contents as compared to the untreated control. Gas liquid chromatography (GLC) of jasmine oil showed increase in Benzyl benzoate, Benzyl acetate, Benzyl alcohol, Eugenol, Transmethyl jasmonate, Cis-jasmone and Jasmine lactone contents, as compared to the control. The analysis of essential oil composition in croton leaves by GLC identified the following major compounds: Myrcene, 1, 8-cineole, α terpineol, sabinine, β ocimene and anethole.

Key words: Allelopathy, *Jasminum grandiflorum*, croton leaves, water extract, essential oil.

Introduction

Jasmine (*Jasminum grandiflorum*), plant with universal appeal and a long history of use as a cosmetic, medicine, perfume and incense by many different cultures around the world. It is a member of a small family known as the Oleaceae. It is known in the East as the "King of Aromatics". Jasmine is the pride of all beautiful gardens. Jasmine plant grows worldwide spreading its fragrance all over. Beautiful jasmine flowers are white in color. It is one of the most beautiful and fragrant amongst all the 15,000 flowering plant species, majorly used as decorative flowers. On the other hand, jasmine posses different medical uses. It has a calming and sedative effect on the nervous system and is especially beneficial in conditions caused by psychological stress and imbalance (Kulkarmi and Ansari, 2004 and Sharma *et al.*, 2005). The leaves of *Jasminum grandiflorum L. possess antioxidant content which has a potential of antiulcer activity (Umamaheswari et al., 2007)*. Jasmine flowers are also used for medical motives and religious purposes. Jasmine oil, which is very popular fragrant oil, contains benzyl acetate, terpinol, jasmone, benzyl benzoate, linalool, several alcohols, and many other compounds.

Weeds are one of the largest problems to plant production worldwide. Weeds affect plant growth and production that may be reduced significantly when weeds compete with them for light, water and minerals (Lehoczky and Reisinger, 2003). So, lack of nutrition may result in poor flowering that is the main production in jasmine shrub. Thus controlling weeds considered an option for increasing flowering production.

Efforts are being made to find out new environment-friendly means of weed management. Using allelopathy for weed management is varietal improvement. Hence, this lead to improved water quality and reduced environmental contamination. Allelopathy refers to the beneficial or harmful effects of one plant on

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another plant. As for example, crop and weed species by the release of chemicals from plant parts by leaching, root exudation, volatilization, residue decomposition and other processes in the agro-ecosystem systems (Rice, 1984 and Lambers *et al*, 1998). Therefore, plant product can be a possible alternative for synthetic herbicides and these may be used as natural herbicides (Mahmood and Cheema, 2004).

Many plant products are found to inhibit germination and growth of plants, which have some herbicides properties (El-Rokiek and Eid, 2009 and El-Rokiek *et al*, 2010a&b).

Research on croton allelopathy has been undertaken to select allelopathic concentration in the field or laboratory. Croton plants are one of the outdoors and indoors ornamental plants grow in Egyptian gardens. Researches on the effect of allelopathic action of different parts of croton plants have been investigated. Sisodia and Siddiqui (2009 and 2010) reported that aqueous extracts of croton leaves and roots inhibited root length and dry weight of *Brassica oleraceae*, *Brassica rapa*, *Melilotus alba*, *Vicia sativa* and *Midicago hispida*.

The objective of this investigation is to explore the effect of croton plant residues on weeds surrounding jasmine trees.

MATERIALS AND METHODS

Laboratory Experiments:

Croton leaves and stems were gathered from Egyptian gardens. A total of 2.5, 5, 10, 15, 20, 25 or 30g of dried leaves and stems (oven dried at 40°C) were transferred into labeled bottles, to which 100 mL of sterile distilled water was added. The mixture was shaken well and allowed to soak for 48h at room temperature and filtered to extracts at 2.5, 5, 10, 15, 20, 25 and 30% concentrations.

To evaluate the allelopathic effect of water soluble extracts of croton leaves and stems, 20 seeds of *Portulaca oleracea* or *Echinochloa colonum* were placed on filter paper Whatman number 3 in Petri dishes. Seven milliliters of the crude extracts at 2.5, 5, 10, 15, 20, 25 and 30% or sterilized distilled water, as a control, were applied to the weed seeds. The Petri dishes were sealed and incubated at 30°C for 10 days. Five replicates were done for each treatment. After five days, 2 ml of the previous extracts was added. The number of germinating seeds was recorded, and the seedlings length of 5 randomly selected germinating seeds was measured. The experiment was repeated twice with one week interval.

Field Experiment:

This experiment was carried out during the two successive seasons 2008 and 2009 for testing croton leaves residue in controlling weeds growing with jasmine trees in Gezayh Village, Imbaba District, Giza Governorate, Egypt. Healthy and nearly uniform jasmine trees grown in a sandy clay loam soil orchard (coarse sand 54%, fine sand 1%, silt 23%, clay 22%, pH 7.5 and E.C. 3.79) at 2x2m apart (1000 trees/feddan [feddan=4200m²]) were selected for the study in this region. Fertilizers were added as calcium super phosphate (300kg/fed), ammonium sulfate (20.6% N) at the rate of 400kg/fed and potassium sulphate (48%, K₂O) at the rate 200kg/fed. Nitrogen and P fertilizers amounts were divided into equal portions and applied monthly starting from March to October during each season, while potassium was applied three times equally from March to September.

Preparation of Material Residues:

Croton leaves were collected from Egyptian gardens and dried in an electric oven at 40°C, then ground into a fine powder.

Croton leaves residues were applied as soil treatments, mixed with soil surface under and around jasmine trees on April 2008 and 2009 as follow:

50g/m², 100g/m², 150g/m², 200g/m², 250g/m² and 300g/m² in addition to untreated trees and hand weeding twice at 30 days interval. Each treatment was replicated three times with complete randomized design.

Data Recorded:

Weed samples from square meter were taken at 60 and 120 days after treatments.

Data on jasmine plants were recorded for each individual tree at the flowering stage, including flower yield/tree and 100 flowers weight. Jasmine concrete percentage and oil yield (kg/tree) were extracted from the picked flowers and were kept under shade at room temperature for the removal of extra moisture and were then subjected to oil extraction.

A-photosynthetic Pigments in Jasmine Leaves:

Chlorophylls a, b and carotenoids were extracted from fresh leaves (mg/g) at flowering stage according to the producer achieved by Saric *et.al.*(1967).

B- Total Carbohydrate Content:

Carbohydrate were extracted from drying finely ground leaves (Powdered) at flowering stage according to Herbert *et al.* (1971), and estimated the total carbohydrate calorimetrically by the phenolsulphoric acid method (Montgomery, 1961).

C- Macro- and Micro-Elements:

Macro and micro elements were determined in dried leaves at flowering stage according to the official and modified methods of analysis (AOAC, 1984).

Determination of Essential Oil:**In Jasmine Flowers:**

Flowers were collected from the experimental area in paper boxes and weighed before and after the removal of unwanted material, and were kept under shade at room temperature for the removal of extra moisture and subjected to oil extraction.

Flowers were placed in vessels and covered with hexane solvent. The solvent gently heated electrically, the solvent extract contain the chemical fragment were transferred to alcohol. Alcohol was removed by low heat distillation. The oil yielded was dried over anhydrous sodium sulphate and stored in sealed vials at low temperature before analysis (Gunther, 1961).

In Croton Leaves:

The fresh leaves (2Kg) of the plant were dried on a laboratory table for 8 days and reduced to powder. The 100g powder was taken for oil extraction with ethanol using Soxhlet apparatus. The extract was stored in a refrigerator at 4 °C until used for analysis.

Chromatographic Volatile Oil Analysis (GC, GC-MS):

A Hewlett Packard model 3400 gas chromatograph equipped with a DB5 capillary column (30x0.25 m i.d 0.25 mdf.), FID. The separated components were identified by matching with NIST mass spectral library data, and by comparison of Kovat,s indices with those of authentic components and with published data (Adams, 1995). The quantitative determination was carried out based on peak area integration. Using helium as the carrier gas with a linear velocity of 31.5 cm s⁻¹, split ratio 1/60, ionization energy 70 eV, scan time 1 sec., transfer line temp. 260 °C, oven temperature programmed.

Physio-chemical Analysis:

Some jasmine oil characters i.e. refractive index at 20 °C, specific gravity at 15 C°, acid number and ester number were determined according to Gunther (1961).

Statistical Analysis:

The obtained data were submitted to standard analysis of variance; the LSD values were obtained when F values are significant at 5% level (Snedecor & Cochran, 1980).

3- Results:**Laboratory Test:****Effect of Water Extracts of Both Croton Leaf and Stem on Early Seedling Growth:**

Water extracts of leaves and stem of croton plants have no effect on germination percentage of weeds (not showed). Seedling length of *Portulaca oleracea* and *Echinochloa colonum* weeds were significantly reduced by extracts of both leaves and stem of croton as compared to the untreated control, but the effect depended on type and concentrations of the extract. Seedling length of both tested weeds was completely inhibited by 30% leaves extract. The inhibition, reached 85 and 70% of the untreated control in *Portulaca oleracea* and *Echinochloa colonum* (Figure 1) stem extract.

Weeds:

The major weed species which were found around jasmine trees were annual broad leaved weeds, i.e. *Amaranthus spp.* and *Portulaca oleracea*; *Convolvulus arvensis* which is a perennial broadleaved weed, while the dominant grass weeds were *Ecchinochloa colonum* (annual), *Cyperus spp.* and *Cynoden dactylon* (perennial).

Results in Table (1) show that croton leaves residue (CLR) reduced the dry weight of broad leaved weeds and grasses significantly as compared to the corresponding unweeded control and the degree of inhibition varied

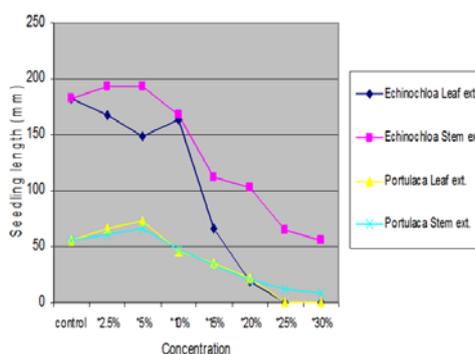


Fig. 1: Effect of different concentrations of croton plant extract on seedling length of *Echinochloa colonum* and *Portulaca oleracea* weeds

between different rates. Using CLR at 300 g/m² caused a significant inhibition of broadleaved weeds after 60 days from treatments reaching to 85% as compared to the unweeded control. The reduction in weed growth continued during the experimental period reaching 87.6 % inhibition. The degree of inhibition in grasses reached 71 and 88.6% at 60 and 120 days after treatments, respectively. So, broadleaved weeds and grasses control ranged from 0 to 88.6% and averaged 87.6 and 88.6% respectively after 120 days with applying CLR at 300g/m². It is interesting to note that even the lowest rate of CLR caused significant reduction in weed growth.

Jasmine:

Flower Yield, Weight of 100 Flowers Concrete Percentage and Oil Yield:

Significant increases were observed in jasmine flower yield, weight of 100 flowers, concrete percentage and oil yield by using CLR from 50-300 g/m². Flower yield as well as weight of 100 flowers was higher when jasmine trees treated with 300 g/m² of CLR. In addition the pattern of the increase in concrete and oil yield was similar.

Table 1: Effect of croton leaves residue (CLR) on dry weight (g/m²) of broad and narrow leaved weeds at 60 and 120 days after treatment (DAT) (Average of the two seasons).

Treatments (g/m ²)	Broadleaved weeds		Grass	
	60 DAT	120 DAT	60 DAT	120DAT
Unweeded	27.5	88.2	30.6	93.2
Hand weeded	10.4	35.7	12.4	49.6
50 CLR	22.4	75.5	27.8	77.5
100 CLR	19.7	67.3	26.0	51.9
150 CLR	16.3	54.1	23.9	41.3
200 CLR	10.9	34.4	20.9	26.0
250 CLR	8.2	26.1	14.6	12.9
300 CLR	4.4	11.0	8.8	10.7
LSD at 5%	1.1	3.9	0.9	3.6

Chlorophyll Contents:

Generally, the application of CLR at all rates induced a significant increase in chlorophyll a & b, total chlorophyll and carotenoid contents over unweeded control, especially with the highest rate (300 g/m²). The values of pigment contents recorded by hand weeding treatment in leaves of jasmine trees exceeded that measured by all rates of CLR, while the least content was measured in leaves of unweeded control (Table 3). It is noted that the increase in chlorophyll pigment was concomitant with increasing in carbohydrate content, consequently, the pattern of change was more or less similar to that of chlorophyll (Table 3).

Macro- and Micronutrient Contents:

The content of N, P, K and Mg in leaves of jasmine treated with different rates of CLR was significantly higher than that measured in unweeded leaves. All recorded higher values especially N that exceeded 200% in leaves of those trees treated with 300 g/m² compared to control. The content of Fe, Mn, Zn and Cu elements in jasmine leaves were significantly increased with all rates of CLR over corresponding unweeded controls. It is interesting to note that Cu content exceeded all measured values of macro and micronutrients (579%) by applying CLR at 300 g/m². However, the content of both macro and micronutrients in leaves of trees in hand weeded treatment exceeded all measured values in comparison to the corresponding controls (Table 4).

Table 2: Effect of croton leaves residue (CLR) on flower yield, weight of 100 flowers concrete percentage and oil yield (Average of the two seasons).

Treatments (g/m ²)	Flower yield kg/tree	Weight of 100 flowers (g)	Concrete %	Oil yield (kg/tree)
Unweeded	0.74	5.11	0.12	0.89
Hand weeded	3.44	13.82	0.36	12.39
50 CLR	0.92	7.22	0.14	1.29
100 CLR	0.93	7.34	0.20	1.86
150 CLR	1.83	9.31	0.25	4.58
200 CLR	2.11	10.21	0.27	5.70
250 CLR	2.92	11.52	0.31	9.05
300 CLR	3.02	12.41	0.32	9.66
LSD at 5%	0.01	0.01	0.02	0.03

Table 3: Effect of croton leaves residue (CLR) on chlorophyll a, chlorophyll b, and total chlorophyll and carotenoid contents (mg/g fresh leaves) in jasmine leaves (Average of the two seasons).

Treatments (g/m ²)	chlorophyll			Carotenoids	Total carbohydrates
	a	b	Total		
Unweeded	1.05	0.431	1.481	0.437	15.41
Hand weeded	2.64	0.752	3.392	0.611	28.14
50 CLR	1.13	0.501	1.631	0.321	17.43
100 CLR	1.35	0.543	1.893	0.532	19.74
150 CLR	1.42	0.632	2.052	0.343	21.45
200 CLR	2.13	0.684	2.814	0.589	22.83
250 CLR	2.25	0.690	2.94	0.521	23.80
300 CLR	2.54	0.714	3.254	0.641	25.90
LSD at 5%	0.01	0.003	0.061	0.002	1.15

Physical and Chemical Properties of Jasmine Oil:

Physical properties of jasmine oil indicated increase in refractive index and specific gravity with different rates of CLR, especially higher ones (Table 5). However, there was regression in acid number and ester number specially when using higher rates as compared to the unweeded control, but less than hand weeding treatment.

Table 4: Effect of croton leaves residue (CLR) on macro and micronutrients content in jasmine leaves (average of the two seasons).

Treatments Kg/m ²	Macronutrients %				Micronutrients (ppm)			
	N	P	K	Mg	Fe	Mn	Zn	Cu
Unweeded	1.93	0.121	2.14	18.5	130.2	32.8	24.2	0.215
Hand weeded	6.34	0.268	5.78	28.7	178.5	56.3	44.7	1.490
50 CLR	1.98	0.125	2.35	21.2	138.6	35.7	27.4	0.289
100 CLR	2.10	0.132	2.46	21.9	144.8	40.2	31.3	1.340
150 CLR	2.35	0.140	3.48	23.8	152.7	46.7	34.7	1.350
200 CLR	3.46	0.145	3.79	26.4	159.3	51.3	36.2	1.380
250 CLR	5.38	0.167	4.76	26.9	167.4	52.8	39.6	1.430
300 CLR	6.12	0.243	5.71	28.1	173.2	54.7	41.2	1.460
LSD at 5%	0.02	0.04	0.10	0.04	1.4	1.0	0.8	0.021

Table 5: Effect of croton leaves residue (CLR) on percentage of major constituents of essential oil in jasmine (average of the two seasons).

Oil properties	Rt	Croton leaves residue (g/m ²)							
		Unwedded	Hand weeded	50 CLR	100 CLR	150 CLR	200 CLR	250 CLR	300 CLR
Physical properties									
Refractive Index	-	1.30	1.42	1.32	1.33	1.35	1.39	1.41	1.42
Specific Gravity	-	0.84	0.849	0.840	0.840	0.843	0.845	0.85	0.84
Ester Number	-	55.42	51.40	49.30	49.70	49.1	45.7	45.20	44.20
Acid Number	-	53.50	46.50	46.20	44.50	43.20	42.40	40.22	40.10
Chemical fraction									
Benzyl benzoate	0.678	22.50	26.20	22.31	22.83	23.44	24.40	24.20	25.50
Benzyl acetate	0.694	9.42	16.40	9.46	10.51	13.34	13.8	13.85	14.10
Benzyl alcohol	0.789	0.80	1.48	0.91	0.92	1.12	1.28	1.24	1.41
Eugenol	0.821	1.85	2.45	1.85	1.81	2.31	2.50	2.20	3.11
Transmethyl jasmonate	0.949	12.80	17.50	14.71	14.83	14.95	16.74	16.10	18.20
Cis-jasmone	1.741	11.83	15.21	12.80	12.87	13.90	15.73	15.21	15.94
Jasmine lactone	1.943	12.47	15.20	13.58	13.50	14.21	14.83	15.20	15.83
Methyl anthranilate	2.198	0.93	1.68	0.94	0.85	1.41	1.41	1.44	1.62

All chemical constituents of jasmine oil (Table 5) of plants treated with CLR were significantly lower than that of hand weeding treatment, however its higher than that of unweeded control. In general, detectable

increases were recorded with higher rates. The increases were recorded especially in Benzyl benzoate, Benzyl acetate, Benzyl alcohol and Transmethyl jasmonate constituents.

4- Discussion:

Continuous use of synthetic herbicides in heavy doses creates environmental pollution and increases the number of herbicide resistant weeds. Hence, researches should be done to find out some natural way for minimizing the dependency on synthetic herbicides. Allelopathy is the inhibitory or stimulatory effect of a plant (donor) on other plants (receivers) through the chemicals released from the donor plant to the environment, mostly into the soil. These chemicals may reach the receiver plants in various ways, including leaching from plant foliage, exudation from the roots, and decomposition of dead residue of the donor plants (Kobayashi, 2004). Hence, the idea of using plant residues.

The results of this study showed that CLR was effective against weeds growing with jasmine trees even with low rates (Table 1). The allelopathic effects of CLR against broadleaved weeds and grasses appeared to be associated with the rate applied. This means that higher rates have higher allelopathic potential; this may be attributed to the relative amount of allelochemicals that was released from the croton residue in the field. Aqueous extract of croton leaves and roots inhibited seedling growth of *Brassica oleraceae*, *Brassica rapa*, *Melilotus alba*, *Vicia sativa* and *Midicago hispida* (Sisodia and Siddiqui, 2009 and 2010). In addition, numerous substances that posse's allelopathic potential has been found in several plant species. For example, benzoic, ferulic, hydroxybenzoic and cinnamic acids were found in the residue of some composite weeds, foliage and tubers of *Cyperus rotundus* and leaves of mango and other plants (Chon *et al.*, 2003; El-Rokiek *et al.*, 2010a&b and Bhadoria, 2011). Some compounds of volatile oils in *Eucalyptus citriodora* leaves inhibited weed growth (Singh *et al.*, 2005; El-Rokiek and Eid, 2009 and El-Rokiek and El-Nagdy, 2011).

Chromatographic analysis of volatile oil from croton leaves contain some terpinoids such as α terpineol and other compound (Figure 2) that may be have a role in inhibiting weed growth. Several workers supporting this suggestion; Kil *et al.* (2000) found that essential oil of *Artemisia lavanduaefolia* contain several compounds similar to the resulted compound in our study and responsible for inhibiting seed germination, shoot and root growth of *Lactaca sativa* and *Artemisia princeps*. Similar results were obtained by Liu *et al.* (2003) and Haibin *et al.* (2009).

On the other hand, higher allelopathic potential of CLR against weeds was accompanied with increase in chlorophyll contents in leaves of jasmine. The yielded materials of jasmine as for example biomass of 100 flowers, concrete increased as well. The results also indicated increase in micro and micronutrient contents in leaves of jasmine at the flowering stage due to allelopathic potential of CLR. This can be explained by reduction in weed/plant competition due to weed growth inhibition. Several workers proved that controlling weed reduced weed/plant competition and consequently, increased product income (Abdelhamid and El-Metwally, 2008, El-Rokiek and Eid, 2009; El-Metwally *et al.* 2010 and El-Rokiek *et al.* 2010b).

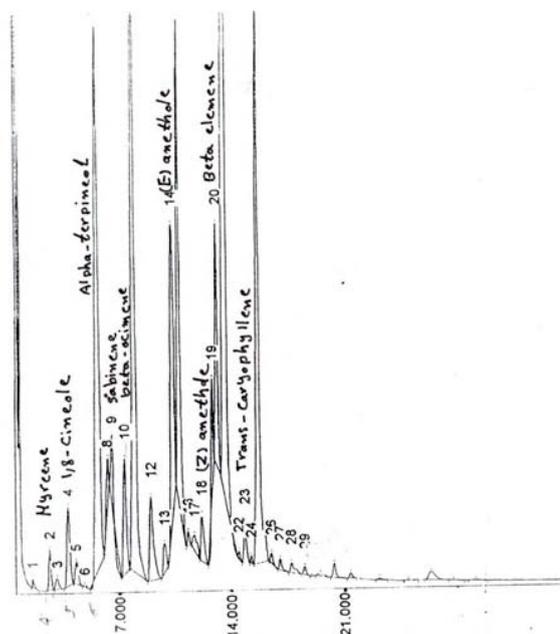


Fig. 2: Chromatogram of oil extracted from croton leaves (*Codiaeum variegatum* L.).

It's important to mention that no treatment recorded more oil yield and its constituents as well as macro and micro elements in plants than hand weeding treatments.

The results also showed improving in quality of jasmine oil due to increase in jasmonoid compounds (Table 5) that characterizes the fragrance of jasmine (Rawia *et al.* 2010). Jasmine oil quality was related with some physical properties as refractive index, specific gravity and acid number. The results indicated, in comparison to the corresponding unweeded control, an increase in refractive index, specific gravity that may be indicated to increasing in unsaturated double bond (iodine value) and decrease in molecular weight (purity) as have been reported by Rudan and Klofutar (1999). However, acid value decrease, the acid value is an indirect measure for free fatty acids that were responsible for unpleasant odor and deteriorate the quality of the product (Mohammad *et al.* 1999).

5- Conclusion:

Croton leaves residue had allelopathy effects on some broadleaved weeds and grasses. GLC analysis of the allelopathic activity of essential oils of croton leaves will be undertaken. This work would be useful for controlling different weeds associated crop plants.

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