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Extract of *Eruca sativa* and Its Effects on Sex Differentiation in Muskmelon

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Authors' contributions

This work was carried out in collaboration between all authors. Authors LAJ, HMH both did the practical work and share ideas for designing the experiments Author NKA designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. All authors read and approved the final manuscript.

Short Communication

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ABSTRACT

The role of herbs in increasing sex capability has been well demonstrated in science, However, the effect of Rocket (*Eruca sativa)* which known, as sex herb in animals, was studied in this work. It was found that 1:10 methanol w/v extract was effective in producing two types of callus white friable callus and compact green callus. The friable callus produce roots while the green callus continues growing without any differentiation.

Keywords: Eruca sativa; cucumismelo; tissue culture; extract.

1. INTRODUCTION

A small group of flowering plants are sexually dimorphic [1], where a population comprises male and female individuals in almost equal proportion (50% each). The female plants of several dioecious species are commercially valued for production of fruits (papaya, kiwi fruit, dates, etc.) and seeds (pistachio, nutmeg, black pepper, jojoba etc.) and therefore, cultivation of female plants in such cases is preferred to the respective male plants. However, as the sex of most of the dioeciously plants is not revealed morphologically, male and female plants cannot be distinguished at the seedling stage. This problem is

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exacerbated in species where the sex of an individual is revealed only after flowering which may take few months (papaya, *Coccinia*) to several years (nutmeg, jojoba).A few dioeciously plants like *Silene* [2], *Rumex* [1], *Humulus* [3] and *Coccinia* [4] possess distinct sex chromosomes and thus the sex of these plants can be distinguished cytologically. However, most of the dioeciously plant species lack such morphologically differentiated sex chromosomes and, therefore, cannot be distinguished based on cytology. To overcome these constraints in cultivation practice and to make it more profitable, there has been a long-standing interest in developing strategies for identifying sex of dioeciously plants at the juvenile stage. In this work it has been demonstrated that genes at the early stages of development can be switched on and off in some *in vitro* treatments. The study of sex ratio in cucurbitaceae has been well established in literature. Researchers found that this ratio depends on: the inheritance factors and some other physiological factors such as temperature, nutrition, photoperiod, and plant growth regulators [5].Regeneration of plantlets is difficult in cucurbitaceous [6] it was found that spray cucumber plants with IAA and NAA increase femaleness while spraying with GA3 increase maleness. However, the effect of Ethaphone has to stop forming male flower buds especially in the lower parts of plants [7].

Eruca sativa or Rocket plant used normally in local markets, this plant has many glycogen level in diabetic's rats. [8] . The antimicrobial activities for rocket leaves has specific importance specially when it is against *E. coli* which is flavonoids activities, alkaloids [9], biofuel [10] and antimicrobial [11,12]. It has been reported that Rocket leaves have some effect on testicular and preputial maturation in male mice [13]. No report has been found on the effect of this plant on dimorphatic sexuality of cucurbitaceae.

2. MATERIAL AND METHOD

Seeds for *Cucumis melo* var. flexuosus were bought from the local market. The seeds were sown in 50 cm pots with normal soil [sandy loam soil]. After the plants grew 100cm in length, the buds less than 2mm were collected from the upper part of plants soaked in 2.5%normal bleach for 5 minutes and washed three times of 5 minutes each time with sterilized distilled water. The sterilized buds were cut vertically with sterilized blade then, cultured in MS medium supplemented with 30g/l sucrose, 2mg/l BA and 1.5 mg/l NAA.

Preparation of extract: The plants of *Eruca sativa* were bought from local markets after dried at room temperature the shoots were then extracted in ten times of methanol (1: 10 w/v) 6-8 hours in a soxculate at 60-80 C, then extractions were filtered through Wattman filters. The residue was dried at room temperature. The powder was used in different concentrations later. The statistical analysis was done according to SAS 2001[14].

3. RESULTS AND DISCUSSION

The results in Table 1 and Figs. 1A, 1B & 1C show that the best concentration for callus induction was 4mg/l extract, However, other concentrations were significantly different from 4mg/l both in dry and fresh weights. These results indicate that the extract concentrations had several effects on callus induction.

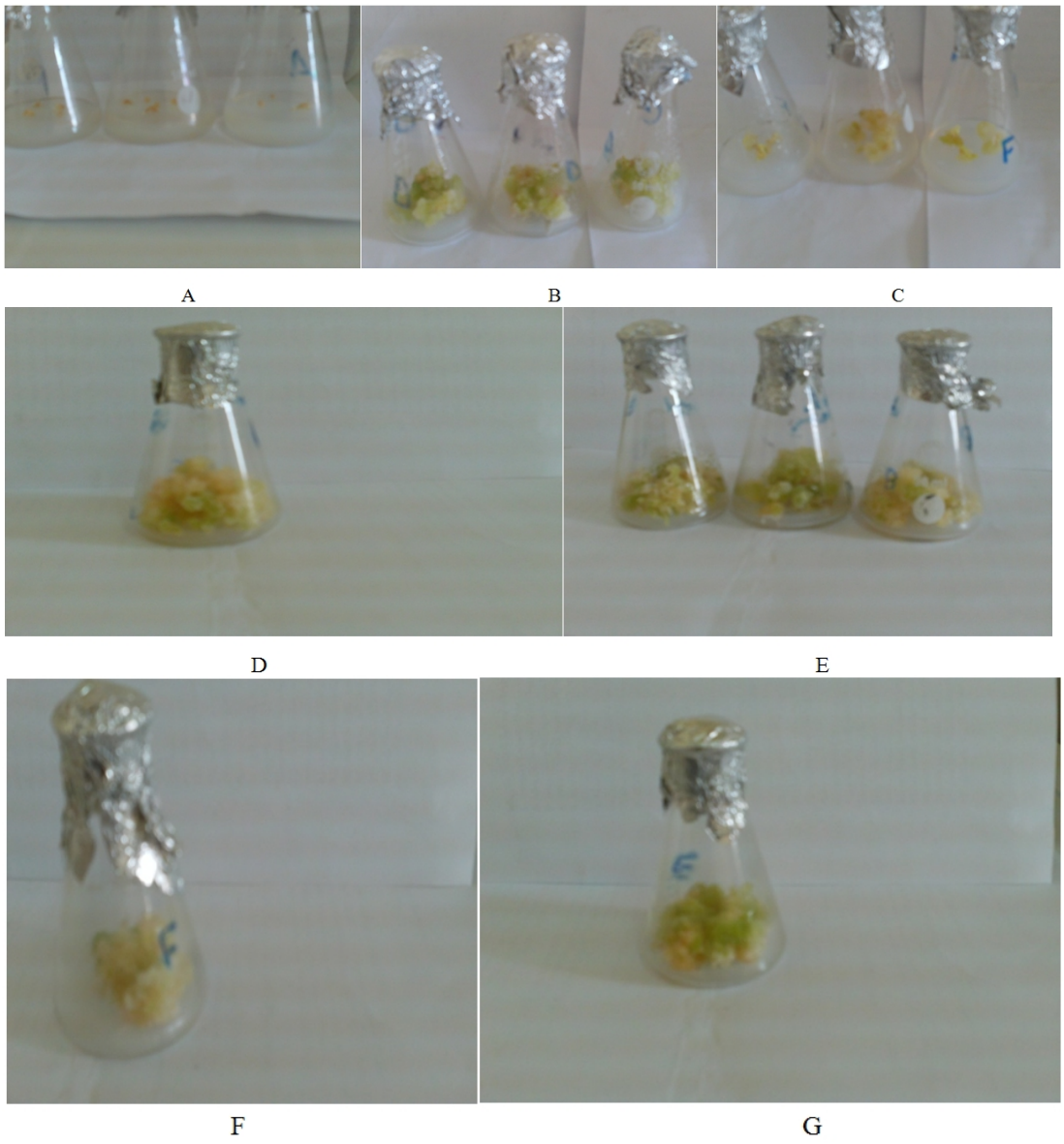

Fig. 1. Callus formation in different stages of the experiment

1. Callus at table 1A. in control B.4mg/l extract C.10 mg/l.
2. Callus at table 2D. white callus origin. E. green callus origin.
3. Callus at table 3F. white callus origin .G. green callus origin.

Table 1. Effect of *Eruca sativa* extract initiation of C*ucumismelo* callus flower buds cultured on MS supplemented with 2 mg/l BAP plus 1.5 mg/l NAA. N = nine replicates

Extract con.	Fresh wt (g)	Dry wt (g)
0+0 hormone	0	0
0+ hormone	6.88	0.38
2+ Hormone	7.85	0.42
4+Hormone	10.06	0.48
6+ hormone	9.11	0.48
8+ hormone	2.80	0.14
10 +Hormone	2.02	0.13
LSD(P =.05)	2.653	0.209

In Table 2 and Figs. 1F & 1G the subculture of the callus obtained from Table 1 was done , but with the separation of white friable callus from green compact callus. It was clear from the Table that white callus gives green and white callus when sub cultured, However, green callus gives greener callus than white. Also the best concentration for that was treatment with 6mg/l extract. Why not 4 ml as before?

Table 2. Subculture of callus from table one with separation of white friable callus and green callus

Extract con. mg/l	White origin callus		Green origin callus		Fresh weight (g)	Dry weight (g)
	White	Green	White	Green		
0.0 +0 hormone	0	0	0	0	00	0
0.0+ hormone	70	30	60	40	9.41	0.25
2.0 + hormone	0	100	0	100	6.2	.20
4.0+ hormone	80	20	0	100	3.71	0.15
6.0+hormone	50	50	30	70	12.82	0.35
8.0 + hormone	70	30	0	100	1.95	0.09
10.0 + hormone	0.0	0.0	0.0	100	7.29	0.21
LSD(P=.05)	11.65	12.75	9.50	12.75	4.59	0.185

The green callus was sub cultured on the same media to regenerate shoots and roots. The results of this experiment are shown in Table 3. In this Table the callus continued growing without any response for shooting even at different concentrations of the extract. This indicates that the extract will not affect the green callus. As we know from previous study that the green callus may be refer to femaleness and the white callus may give maleness [15]. This reflected our attention to the possibility that the extract may only work in male tendency buds.

The white callus was also sub-cultured with different concentrations of the extract. The results in Table 4 indicate the regeneration of roots with different number and length. The best response was on 4mg/l extract and hormone. These results will give us a clear response for male tendency callus (white callus) to the extract treatment when we compare the results in Table 3 and 4 are compared.

Table 3. Effect of extract on green callus regeneration

Extract con mg/l	Responses	Fresh wt (g)	Dry weight (g)
0.0 +0 hormone	0.0	0.0	0.0
0.0+ hormone	0.0	23.13	0.95
2.0 + hormone	0.0	29.88	0.25
4.0+ hormone	0.0	21.25	0.76
6.0+hormone	0.0	20.24	0.96
8.0 + hormone	0.0	18.35	0.84
10.0 + hormone	0.0	14.42	0.57
LSD(P=.05)	0.0NS	6.595	0.362

Table 4. Effect of different concentrations of extract on white callus cultured on 1.5mg/l NAA

Extract con. mg/l	Number of roots	Root length(CM)	Fresh weight (g)	Dry weight (g)
0.0 +0 hormone	0.0	0.0	0.0	0.0
0.0+ hormone	49	0.20	7.25	0.30
2.0 + hormone	12	1.0	2.21	0.16
4.0+ hormone	4	1.25	9.77	0.63
6.0+hormone	40	1.0	14.39	0.73
8.0 + hormone	0.0	0.0	4.23	0.31
10.0 + hormone	0.0	0.0	0.0	0.0
LSD(P=.05)	6.47	0.250	4.208	0.266

4. CONCLUSION

In a conclusion the extract may affect the white callus more than green callus even though both were imitated from upper part of the plant which has a tendency of producing female flowers.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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