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The Antigenotoxicity of *Eruca sativa* Mill Extract on Bone marrow Cells of Male Albino Mice Treated With Vincristine

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Abstract

Chemotherapy drugs have a major role in fighting cancer but, that doesn't mean they are clear from side effect on human health. One of these drugs is vincristine this study, the chemopreventive potency of the crude extract ofIn Eruca Sativa Mill Which have a good reputation as antioxidant and anticancer against chromosomal aberration and suppressed mitotic index in bonemarrow cells of male albino mice treated with 50µl of vincristine intrapritoneally(ip) have been detected.

Results Showed a Significant increasing in the percentage of the mitotic index nearly to the control group and a significant decrease in chromosomal aberration after (35) days of treatment with 250 mg/kg crude extract of *Erucasativa*. These results suggested a strong antigenotoxicity effect against vincristine damages onbonemarrow cells.

Key Words: Antigenotoxicity ,Bonemarrow cell, Vincristine ,*Eruca Sativa*

Introduction

Folk medicine uses many medicinal herbs, Which are not free from Toxic effects. Some of them can cause adverse effects or interact with other medication [1]. It is Known that green plants in general are primary source of antimutagens as Well as a natural toxic agent can be considered as mutagenic, clastogenic and carcinogenic both*in vivo* and [1,2,3,4,5]. *troIn vi*

It this study the extract of *Eruca sativa* Mill Which commonly Known as Rocket plant or Aljarjeer was used .It is a herbaceous plant with great importance as salad vegetable and spice, especially among Mediterranean people[6]. It is one of Brassicacae family possessesmedicinal and therapeutic properities including inhibition of tumorigenesis[7], eye infection, digestive and Kidneyproblems,antiscrbutic,diuretic and it's always considered potent aphrodisiac [6]. Also, it has been reported the antioxidant and anticancerabilities [8].Another research showed the cytoprotective and antiulcer activities[9]

Lamy*et al.* [10] showed the antigenotoxicity of rocket extract on (HepG2) cells (human hepatoma cells) . Non aqueous extract of this plant showed maximum antibacterial activity against BLB bacterial leaf blight disease of rice[11]. Also, antimicrobial resistant gram negativity and positive bacteria [12].Methanolic extract of rocket enhanced the activity of clarithromycin against resistant *E.coli*strains [13].Finally,the rocket seed extract was used as anti-inflammatory[14].

Material and Methods

A-*Eruca sativa* extraction**:**

The plant material was purchased from local market in Baghdad, Iraq. For the extraction, the plant was first washed free of sand ,then crushed in to very small piecesand the juice was subseguently squeezed out[10].

B-Animal treatment:

Twenty four adult male mice with weight rang between(30-36) gm were purchased form Biotechnology Research Center and maintained on (10-14) hour light dark cycle in the animal house. Control and treated mice were provided with food and water *ad libitum*, there were no differences in food intaka, each composed of eight mice.

The first group received normal saline as control group. The second group received 50µ1 of vincristine obtained from BDH chemicals(i.p) intrapritoneally.

The third group received the same does of vincristine then treated

With 250 mg/kg of body weight crude extract of *Eruca sativa* for35 days of treatment.

C-Cytogenic experiments:

1-Chromosome preparation from mouse bonemarrow. This preparation was done according to Allen*et al.*[15].

2-Mitotic Index (M.I)

The slides were examined under high dry power (40X) of light microscope and (1000) divided and non-divided cells were counted and the percentage rate was calculated for only the divided ones according to the following equation:-

$$M.I = \frac{\text{No.ofdividing cells}}{\text{No.ofdividingcells+No.ofnondivding cells}} \times 100[16]$$

D- Statistical analysis:

Statistical analysis was performed using (ANOVA) test. Statisticalsignificancewas determined at $p<0.05$[17]

Results

A-The effect of*Eruca sativa* extract on mitoticindex in bonemarrow cells of male albion mice treated with vincristine:

Table (1) showed a significant ($p<0.05$) decrease in mitotic index of albino mice bonemarrow treated with vincrstine but, after the use of*Eruca sativa* extract there was an increase in the percentage of mitotic index similar to that found in control group.

B-The effect of *Eruca sativa* extract on chromosomal aberration induced by vincristine in bonemarrow cells of male albino mice:

There was significant increase ($p<0.05$) in the percentage of chromosome aberration compared with control group Table(2), the percentage of ring chromosome, was similar to that found in control. Due to the use of*Eruca sativa* extract, obvious decrease in the percentage of chromosome aberration was found.

Discussion

In spite of the benefits of chemotherapeutic drugs against cancer. It also, can cause chromosomal abnormalities due to the interfering with cell division and targeting the synthesis and function of DNA by binding to it (18). They considered a clastogenic compound [19, 20]. Also, they can affectct the fertility and progeny outcome [12].

Vincristine is one if these drugs which used as anticarcinogenic.Various types of secondary tumors in normal cells could resulting from prolonged use of these drugs . So, supplementation with natural antioxidant material may be reduced thegenotoxicity caused by chemotherapy [22, 23]. Due to the good reputation of *Eruca sativa or* rocket plant or aljarjeer

which is widely used in folk medicine ;this study tried to evaluate the effect of this plant extract on the genotoxicity caused by vincristine .the results in table 1 and 2 showed a significant arising in mitotic index percentage and a significant decrease of chromosome abnormalities and that is similar to the results of Lamy*et al.*[10] when they noticed the antigenotoxicity of rocket extract on human hepatoma (HepG2) cells towards benzo (a) pyrene induced DNA damage. Many researches explained the antimutagenic potential of different plant extracts[24,25,26,23].

Concerning the results of this study, the putative role of*Eruca sativa* extract on cancer chemoprevention due to the bioactivity of (GLS) Glucosinolate hydrolysis product namely (Isothiocynate) ITCs and its derived Erucin (ER) which is a major component found in rocket salad leaves and other parts.

There is so manyevidenes about the cancer chemoperevention of ER *in vitro* and *in vivo* studies [8]. The antioxidant effect of ER due to it's reactivity with hydrogen peroxide and alkylhydroperoxides accumulated ln cell and peripheral blood to form water and an alcohol [27,28], or induction of phase II enzyme, or acting as precursor of sulforaphene (potent inducers detoxification electrophiles and increase cellular antioxidant defenses) [28,29].

Other compounds beside ITCs may also protect DNA from damage, like: polyphenolic compounds, carotenoids, volatile oils, flavonoids [10,14,30,31,32,33,34]. Vitamin C which also antioxidant both *in vitro* and *in vivo* has the potential of altering the genotoxic effects of reactive oxygen metabolic (ROMS) which can promote carcinogenesis by damaging DNA [35,36]. All these compounds known for theirchemopreventive activities and the interaction of these compounds led to the strong antigenotoxicity of *Eruca sativa* extract [10].

References

1- Zink, T. and Chaffin, J. (1998). Herbal health products: What family physicians need to know. American Family Physician, 58(5):1133-1140

2- Higashimoto, M.; Purintrapiban, J. and Kataoka, K. (1993).
Mutagenicity and antimutagenicity of extract of three species and medicinal plant in Thailand.Mut. Res., 303(3): 135-142.

3- Plewa, M. J. and Wagner, E. D. (1993). Activation of proutagens by green plants. Ann. Rev of Genetics, 27: 93-113.

4- Nandi, P.; Talukader, G. and Sharma, A. (1998). Plants against cancersome aspects. Nucleus, 41(1-2): 53-86.

5- Aşkin-Çelik, T. and Aslantürk, S. (2007). Cytotoxic and genotoxic effects of *Lavandulastoechas*aqueous extracts.Biologia, 62(3): 292-296.

6- Varga, J.; Apahidean, A. S.; Lujerdean, A. and Bunea, A. (2009).
Study of some agrotechnological characteristics of rocket *(Eruca sativa* Mill) Bulletin USAVM Horticulture, 66(1): 472-474.

7- Lynn, A.; Collins, A.; Fuller, Z.; Hillman, K. and Ratcliff, B. (2006).
Cruciferous vegetables and colo-rectal cancer. Proc. Nutr. Soc., 65: 135-144.

8- Melchini, A. and Traka, M. (2010). Biological profile of erucin: a new promising anticancer agent from cruciferous vegetables. Toxins, 2: 593-612.

9- Al-Qasoumi, S.; Al-Sohaibani, M; Al-Howiriny, T.; Al- Yahyam, M. and Rafatullah, S. (2009). Rocket " *Eruca sativa"* a salad herb with potential gastric anti-ulcer activity. World J. Gastroenterol., 15(16): 1958-1965.

10- Lamy, E.; Schroder, J.; Paulus, S., Brenk, P.; Stahl, T. and MerschSundermann,V. (2008). Antigenotoxic properties of *Eruca sativa* (Rocket plant), erucin and erysolin in human hepatoma (HepG2) cells towards benzo (a) pyrene and their mode of action.Food and Chern.Toxico., 46:2415-2421.

11- Jabeen, R. (2006). Evaluation of botanicals against xanthomonasoryzaepv. Oryzae

causing bacterial leaf blight disease of rice and characterization of bioactive compounds.PHD thesis, Quaid-i-Azam University, Islamabad.

12- Khoobchandani, M.; Ojeswi, B. K.; Ganesh, N.; Srivastava, M. M.; Gabbanini, S., Matera, R.; lori, R. and Valgimigli, L. (2010).Antimicrobial properties and analytical profile of traditional *Eruca sativa* seed oil: Comparision with various aerial and root plant extract. Food Chern., 120 (1): 217-224.

13- Darwish, R. M. and Aburjai, T A. (2010). Effect of ethanomedicinal plants used in folklore medicine in Jordan as antibiotic resistant inhibitors on *Escherichia coli.* BMC complement.Altern.Med., (Abstract).

14- Yehuda, H.; Khatib, S.; Sussan,I; Musa, R.; Vaya, J. and Tami, R.S. (2009). Potential skin anti-inflammatory effects of 4methylthiobutyl isothiocyanate (MTBI) isolated from rocket *(Eruca sativa)* seeds. Biofactors, 35(3): 295-305.

15- Allen, J. W.; Shuler, C.F.; Mendes, R.W. and Latt, S.A. (1977). Asimplified technique for *in vivo* analysis of sister chromatic exchange using 5-bromodeoxy-uridine tablets. Cytogenetics, 18: 231-237.

16- Shubber, E. K. and Juma, A. S. (1999). Cytogentic effects of plant extract of *Urticadioca*on mouse somatic cells. The Nucleus. 42 (3):182-187.

17- Al-Mohammed, N. T; Al-Rawi, K. M.; Younis, M. A. and AIMorani, W. K. (1986). Principle of statistics. J. Al-MosulUniversity (in Arabic).

18- Xiaogang, Q.; John, O.; Trent, I.; Waldemar, P. and Jonathan, B. (2000).Allosteric chiral selective drug binding to DNA. Proc. Natl. Acad. Sci., 97(22): 12032-12037.

19- Arni, P. and Hertner, T (1997). Chromosomal aberrations *in vitro* induced by aneugens. Mutation Research / Fundamental and Molecular Mechanisms of Mutagenesis, 379(1): 83-93.

20- Hongping, D.; Jianlin, L.; Meibian, Z.; Wei, W.; Lifen, J.; Shijie, C.; Wei, Z.; Baohong, W. and Jiliang, H. (2006). Detecting the cytogenetic effects in workers occupationally exposed to vincristine with four genetic tests. Mutation Research/ Fundamental and Molecular Mechanisms of Mutagenesis, 599(1-2): 152-159.

21- Vaisheva, F.; Delbes, G.; Hales, B. and Robaire, B. (2007). Effect of the chemotherapeutic agents for non-Hodgkin lymphoma, cyclophosphamide, Doxorubicin, Vincristine and Prednisone (CHOP), on the male rat reproductive system and progeny outcome. J. Andro., 28(4): 1-17.

22- Selim, N. A.; Abdel-Khalek, A. M., Nada, S. A.and EI-Medany, Sh.A. (2008). Response of growing rabbits to dietary antioxidant vitamins E and C. 1- Effect on performance. Nutrition and digestive physiology In: Proc. 9th world Rabbit congress. Verona, Italy: 803-807.

23- Hassanane, M. M.; Ahmed, E. S.; Shoman, Th. M. and Ezz-Eldin, A. (2010). Evaluation of genotoxicity and antigenotoxicity of curcumin by chromosomal aberration and biochemical studies in the albino rats exposed to Methotrexate. Global Veterinaria, 4(2): 185-189.

24- El-Ashmawy, 1. M.; El-Nahas, A. F. and Salama, O. M. (2005).Basic Clin.Pharmacol.Toxicol., 97: 238-243.

25- Qari, S. (2008). *In vitro* evaluation of the antimutagenic effect *Origanummajorana*extract on the meristematic root cells of *Viciafaba*. J. Taibah Univ. Sci., 1: 6-11.

26- Mohammed, B.; Karim, K. and Yaseen, N. (2009). Antimutagenic effect CTS of *Thymus syriacus*extract against the genotoxicity of gemcitabine in male albino mice. J. Duhok Univ., special issue12(1): 216-226.

27- lori, R.; Bernardi, R.; Gueyrard, D.; Rollin, P. and Palmieri, S. (1999). Formation of glucoraphanin by chemoselective oxidation of natural glucoerucin: achemo-enzymatic route to sulforaphane. Bioorg. Med. Cherm. Lett., 9: 1047-1048.

28- Barillari, J.; Canistro, D.; Paolini, M.; Ferroni, F.; Pedulli, G.; Lori, R. and Valgimigli, L. (2005). Direct antioxidant activity of purified glucoerucin the dietary secondary metabolite contained in rocket *(Eruca sativa* Mill) seeds and sprouts. J. Agric. Food Cherm., 53(7):2475-

2482.
29- El-Nattat, W. S. and El-Kady, R. 1. (2007). Effect of different medicinal plants seeds residues on the nutritional and reproductive performance of adult male rabbits. lnternat. J. Agric. Bio., 9(3): 479-485.
30- Jirovetz, L.; Smith, D. and Bnchbauer, G. (2002). Aroma compound analysis of *Eruca sativa* (Brassicaceae) spme headspace leaf samples using GC, GC-MS and olfactmertry. J. Agric. Cherm., 50(16): 4643-4646.
31- Abdo, M. A.; Zeinab (2003). Using Egyptian *Eruca sativa* seed meal in broilerration with or without microbial phytase.Egypt J. Nutr.Feed, 6:97-114.
32- Alam, M.; Kaur, G.; Jabber, Z.; Javed, K. and Athar, M. (2007). *Eruca sativa* seeds possess antioxidant activity and exert aprotective effect on mercuric chloride induced renal toxicity. Food Chern.Toxic., 45(6): 910-920.
33- Martinez-Sanchez, A.; Llorach, R.; Gil, M. I. and Ferreres, F. (2007).Identification of new flavonoid glycosides and flavonoid profiles to characterize rocket leafy salads *(Eruca sativa* and *Diplotaxistenuifolia).* J. Agric. Food Cherm., 55(4): 1356-1363.
34- Blazevic, I. and Mastelic, J. (2008). Free and bound volatiles of rocket *(Eruca sativa* Mill).Flavour and fragrance J., 23(4): 278285.
35- Crott, J. W. and Fenech, M. (1999). Effect of vitamin C supplementation on chromosome damage, apoptosis and necrosis *ex vivo*.Carcinogenesis, 20(6): 1035-1041.
36- Rahman, K. (2007). Studies on free radicals, antioxidants, and cofactors.Clin.lnterv. Aging., 2: 219-236 .

Table (1): Percentage of mitotic index in male mice bonemarrow cells of different experimental groups.

Groups	Mitotic index % (mean ± SE)
Control	**A** **10.66 + 1.93**
Vincristne (50µ1)	**B** **3.83 + 1.45**
(250mg/kg) *Eruca sativa* extract	**A** **11.92+ 2.03**

Different letters (A,B,C) mean Significant at (p<0.05)

Table (2): Percentage of chromosomal aberration inmale mice bone

Groups	Chromosomal Aberration % m± SE								
	Chromatid break	Chromatid gap	Deletion	Dicentric	Acenrtric	Ring	Chromosome break	Chromosomegap	Total
Control	A 0.05+0.005	A 0.04+0.006	A 0.19+0.06	A 0.18+0.009	A 0.21+0.006	A 0.04+0.008	A 0.08+0.04	A 0.02+0.009	A 0.81+0.023
Vincristine (50µ1)	B 0.22+0.09	B 0.21+0.07	B 0.32+0.07	B 0.51+0.32	B 0.82+0.33	A 0.04+0.004	B 0.12+0.006	B 0.14+0.30	B 2.38+0.81
(250mg/kg) *Eruca sativa* extract	C 0.00+0.00	A 0.01+0.003	A 0.12+0.08	C 0.03+0.01	C 0.06+0.05	B 0.01+0.003	A 0.03+0.008	A 0.00+0.00	A 0.35+0.07

marrow of different experimental groups
Different letters (A,B,C) mean significant at ($p<0.05$)

الفعالية المضادة للسمية الجينية لمستخلص نبات الجرجير على خلايا نقي العظم لذكور الفئران البيض المعاملة بعقار الفنكرسيتن

شيماء صباح مهدي
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الخلاصة

العقاقير المضادة للأورام لها دور كبير وأساسي في محاربة السرطان. لكن هذا لايعني خلو هذه العقاقير من التأثيرات الجانبية في صحة الإنسان، احد هذه العقاقير هو الفنكرستين. في هذه الدراسة تم التحقق من القابلية الوقائية للمستخلص الخام لنبات الجرجير والذي يحظي بسمعة جيدة بوصفهنباتاً مضاداً للسرطان وضد التشوهات الكروموسومية وانخفاض دالة الانقسام لخلايا نقي العظم في ذكور الفئران البيض المعاملة بجرعة 50 مايكروليتر من الفنكرستين المحقون داخل الخلب البريتوني.

بينت النتائج زيادة معنوية في دالة الانقسام مقاربة لنسبة مجموعة السيطرة وكذلك انخفاض معنوي للتشوهات الكروموسومية بعد خمسة وثلاثين يوما من المعاملة بجرعة 250غم لكل كيلو غرام من وزن الجسم بالمستخلص الخام من نبات الجرجير. هذه النتائج تبين تأثيرا مضاداً للسمية الجينية في خلايا نقي العظام الناتجة من المعاملة بعقار الفنكرستين.

الكلمات المفتاحية : الفعالية المضادة للسمية الجنية ،خلايا نقي العظم، عقار الفنكرستين ،الجرجير .