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EUPHORBIA HELIOSCOPIA: CHEMICAL CONSTITUENTS AND BIOLOGICAL ACTIVITIES

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ABSTRACT

Euphorbia, the largest genus in the spurge family “Euphorbiaceae” with more than 2000 species and is subdivided into many subgenera and sections. Several species of the genus *Euphorbia* have been extensively studied for their antiviral, antitumor, cytotoxic, antimicrobial and pesticidal activities. Based on traditional information, *Euphorbia helioscopia* has been widely used in the traditional folk medicine in China and Turkey. Up to now, 30 diterpenoids have been isolated and structurally characterized from this plant. The aim of the present work is to review all the available scientific literatures published on *E. helioscopia*. The focus will be on the chemical constitutions that have been identified from this species, in addition, all the reported biological, pharmacological and toxicological activities of different extracts and isolates from this species have been included. The paper recommends the need for further investigations regarding the environmental and mammalian safety of *E. helioscopia* for safer using in different biological and therapeutic applications.

Keywords: *Euphorbia helioscopia,* chemical constituents, terpenes, flavonoids, volatile oil, biological activities.

INTRODUCTION

Euphorbiaceae is the largest family among the Anthophyta, with 300 genera and 5000 species. The genuses sub cosmopolitan but with strong representation in the humid tropics and subtropics of both hemispheres (Uzair M *et al*., 2009). The genus *Euphorbia* is the largest genus in the Euphorbiaceae family with over 2000 species ranging from annuals to trees and is subdivided into many subgenera and sections. All contain latex and have unique flower structures (Barla A *et al*., 2006; Chaudhry BA *et al*., 2001; Jassbi AR *et al*., 2006). *Euphorbia* species are used for the treatment of various ailments such as skin disease, gonorrhea, migraines, intestinal parasites and

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warts cures. The plant lattices have been used in fish poisons, and insecticide (Uzair M *et al*., 2009). Based on traditional information, the leaves and the lattices of this genus are used in the ayurvedic system of medicine for bronchitis and rheumatism (Barla *et al*., 2006). Furthermore, it is stated to possess inflammatory, antiarthritic, antiamoebic, spasmolytic, antiviral, hepatoprotective and antitumor activities. For hundreds of years with traditional Chinese medicine *Euphorbia* have been used for the treatment of cancers, tumors and warts, and is well known that this pecies contains irritant and tumor-promoting constituents (Yang ZS *et al*., 2009). Quite a number of species are used in folk medicine as drugs and raw materials for pharmaceutical preparations. In Turkish folk medicine, *Euphorbia* species have been used for rheumatism, swelling and especially as a wart remover. However, it can also trigger causes inflammation and diarrhea (Barla, 2006).

Euphorbia helioscopia linn., known also as Lun spurge (known in English as wolf's milk, in French as turnsol), is widely distributed in China, The whole plant has great medicinal importance, often used to treat ascites, edema, pulmonary tuberculosis, tinea and cervical tuberculous lymphaden (Feng WS *et al*., 2009; Feng WS *et al*., 2010; Li, 2007). The leaves and stems are used as febrifuge and vermifuge. The oil from the seeds has purgative properties, the roots are used as anthelmintic and the seeds mixed with roasted pepper have been used in the treatment of cholera (Uzair M *et al*., 2009). Based on some ethnobotanical surveys for medicinal plants used traditionally in different countries, it has been recorded that, *E. helioscopia* is used by local people in Pakistan as cathoratic, antihelminthic and purgative (Qureshi RA *et al*., 2007). In addition, the milky juice from the leaves and fresh stems are used to release pus (Ahmed S *et al*., 2006). In Jordan, milky juice has been used as an antiscorbutic as well as a diaphoretic (Al-Qura'n, 2009) also known as toxic species that cause diarrhoea, general fatigue, dysentery, dizziness, and anoxia (Al-Qura'n, 2005). In China, *E. helioscopia* has been used as a traditional folk medicine for the treatment of malaria, bacillary dysentery and osteomyelitis (Lu ZQ *et al*., 2008). A weed used by early dyers for bluish purple (basic) and red (acid) shades, it acts as an indicator, giving colors resembling litmus, and was probably used at one time as a food colorant (Mell CD, 1927).

Botanical aspects

Morphology

E. helioscopia is a smooth annual plant with an erect, stout stem from eight to twelve inches high, often branched from the base. The branches, as well as the main stem, end in a more or less compound umbel which is subtended by a circle of leaflets. The leaves are scattered along the stem; they are somewhat oblong or wedge-shaped, sometimes nearly round, from one-half to four inches long, finely saw-edged, and narrowed to a short stalk. The rather inconspicuous flowers are of two kinds, the staminate and pistillate on the same plant, both included in a cup-shaped involucre resembling a calyx or corolla. The staminate flowers are numerous, lining the inside of the cup, each consisting of one single stamen in the axil of a very little bract. The pistillate flower is solitary in the centre of the cup and consists of a three-lobed, three-celled ovary which soon protrudes on a long stalk and hangs over the brim of the cup-like involucre. The seeds are reddish-brown, strongly honeycombed. The plant is in bloom from June till October (Fyles F, 1919).

Taxonomy

Euphorbia helioscopia linn is classified into Kingdom: Plantae, Subkingdom: Tracheobionta, Superdivision: Spermatophyta, Division: Magnoliophyta, Class: Magnoliopsida, Subclass: Rosidae, Order: Malpighiales, Family: Euphorbiaceae, Subfamily: Euphorbioideae, Tribe: Euphorbieae, Subtribe: Euphorbiinae, Genus: *Euphorbia*, and species: *Euphorbia helioscopia* Linn. (Doe J, 2010).

Distribution

The plant is native to the temperate regions of Eurasia but has adapted to subtropical conditions. It occurs as high as 3,000 m in India and Pakistan and is found to lat 69° N in Europe and Canada. It behaves as a winter annual in Japan, flowering from April to May. In India, plants flower from December to April on the plains and in May in hilly regions. It is often associated with light textured soils (Holm L *et al*., 1997), and in Upper Egypt it was recorded in many cultivated crops (Mahmoud FM, 1996).

Phytochemistry

E. helioscopia L. has been intensively investigated. Different kinds of secondary metabolites, such as triterpenoids, diterpenoids, flavonoids, tannins and lipids have been isolated from this species by several groups during the past four decades (Durrani AA *et al*., 1967; Zhang W *et al*., 2006).

Diterpenes

Macrocyclic diterpenes

The metabolic pattern of *Euphorbia helioscopia* is heavily characterized by a series of complex macrocyclic diterpenes, (*e.g*. jatrophon, jatrophane, and lathyrane).

Jatrophon type diterpenoids

More than thirty jatrophon type diterpenes have been isolated and structurally characterized from the Japanese *E. helioscopia* L., Few studies on the methanol extract of the fresh leaves and roots of *E. helioscopia*, collected in Kanagawa, Japan have been made in the course of searching for physiologically active substances. Yamamura (1981) have isolated two euphoscopin-diterpene types which have been identified as euphoscopin A (1) and B (2), further investigation by Shizuri (1983 a) and Ohba (1983) resulted in the isolation of three new diterpenes, identified as helioscopinolide A (3), B (4) and C (5). Two new toxic diterpenes euphohelioscopin A (6) and B (7) together with two euphoscopin-type skeleton euphoscopins C (8) and D (9) have also been isolated by Shizuri (1983 b). In connection with highly oxygenated diterpenes that have antitumor activity or promote cancer development in tumor formation, three new diterpenes have been identified: euphornins A (10), B (11) and C (12) as well as, five new diterpenes euphohelins A-E (13-17) isolated by (Shizuri, 1984; Koemura, 1985). Examination of the toxic diterpenes afforded eight new jatrophon type diterpene euphornin D-K (18-25) and twelve new euphoscopin-type diterpene: euphoscopin E-L (26-33) and epieuphoscopin A (34), B (35), D (36) and F (37) and euphohelionon (38) (Yamamura S *et al*., 1989). Up to now only two obvious cytotoxic macrocyclic diterpenoids ester have been reported from this plant during the past decades: euphornin (39) (Lu, 2008; Jassbi, 2006) and euphornin L (40) (Tao, 2008). Three new diterpene analogues, euphoheliosnoids A-C (41-43), have been isolated from *E. helioscopia* L. which collected in Shujiae, Zhejiang province, China; all of these new compounds demonstrate considerable spectral analogy with the previously reported euphoscopins but they are either esterified differently at C-7 or oxidized with accompanying migration of a double bond at C-11 (Zhang W *et al*., 2005). Additionally investigation of this plant collected from Zhejiang province has led to the isolation of a new diterpenes with a jatrophon type skeleton, named euphoheliosnoid D (44) (Zhang W *et al*., 2006).

Jatrophane-type diterpenes

From the aerial parts of *Euphorbia helioscopia*, collected from Istanbul, Turkey, a jatrophane diterpene ester, 5,11-jatrophadiene-3- benzoyloxy-7,9,14-tri-acetyloxy-15-ol (**45**) (Barla A *et al*., 2006). Other novel diterpenes were isolated and identified as jatrophane skeleton type, named euphoscopin M (46), euphoscopin N (47) and euphornin L (48) were isolated from the whole plant collected at the Saepinum ruins, in Altilia, Italy (Barile E *et al*., 2008; Corea G *et al*., 2009). From the 95% EtOH extract of the whole plant of *E. helioscopia* collected in Sichuau, China, new jatrophane type diterpene was isolated and identified as euphornin N (49) (Geng D *et al*., 2010). From the 95% EtOH extract of the whole plant collected from Xuzhou, Jiangsu province, four jatrophane type diterpenoids were isolated and identified as 7β,9α,14β-triacetoxy-3β-benzoyloxy-12β,15β-epoxy-11β-hydroxy-jatropha-5E-ene(50),14β-Acetoxy-3β-benzoyloxy-7β,9α,15β trihydroxyjatropha-5E,11E-diene (51), 7β,9α,14β-triacetoxy-3β-benzoyloxy-15β,17-dihydroxy-jatropha-5E,11E-diene (52), 14α,15β-diacetoxy-3β,7β-dibenzoyloxy-17-hydroxy-9-oxo-2βH-jatropha -5E,11E-diene (53) (Lu ZQ *et al*., 2008).

Lathyrane diterpenes

From the 30% MeOH extract of the whole plant of *E. helioscopia* L., collected from Xixia county of Henan province, a new lathyrane diterpenes glycoside has been isolated and identified as 3β,7β,15β-trihydroxy-14-oxolathyra-5E,12E-dienyl-16-O-β-D-glucopyranoside (54) (Feng, 2010). The lathyrane diterpene euphohelioscopin C (55) was isolated from the whole plants, collected at Altilia (Barile E *et al*., 2008; Corea G *et al*., 2009).

Triterpenes

Triterpenoids resembling lupeol were isolated from the latex of Turkish *E. helioscopia* L. with structures confirmed as 19αH-lupeol (**56**) (Nazir, 1998); lup-20(29)-ene-3-acetate (**57**) and lup-20(29)-ene-3-palmitate (58); together with common triterpenoids, were also found and identified as: 24-methylene cycloartanol (59), 24-methylene cycloart-3-one (60), cycloartanol (61) and stigmast-4-ene-3-one (62) (Barla A *et al*., 2006).

Flavonoids

Flavonoids are popular compounds for chemotaxonomic surveys of plant genera and families. Several studies indicated that flavonoids occur in *E. helioscopia* Linn. The qualitative composition of flavonoids in alcoholic extract of *E. helioscopia* indicated 15 substances with flavonoidal nature. By 2-dimensional paper chromatography (2-DPC) after acid hydrolysis of *E. helioscopia* alcoholic extract by Volobuevra (1970) and Abd-Salam (1975) two compounds were characterized: quercetin and kaempferal. *E. helioscopia* appears to have low flavonoids verity compared with other Euphorbia species In the leaves, flavonoid sulphate and flavone C and C-*O*- glycosides) (Noori M *et al*., 2009; Aqueveque P *et al*., 1999) have been reported. Quercetin-3-β-glucoside, quercetin-3-β-galactoside and quercetin-3-β-galactoside-2``-galate were isolated from *E. helioscopia* (Pohl R *et al*., 1975).

Tannins

E. helioscopia L., unlike other *Euphorbia* series so far examined contains large numbers of novel ellagitannins. A study by Lee (1990) reports the isolation of four hydrolysable tannins named helioscpinins A and B and helioscopins A and B having a variety of phenolcarboxylic acid ester groups. Further chemical study on tannins of this plant has led to the isolation of two minor hydrolysable tannins named euphorscopin and euphorhelin (Lee SH *et al*., 1991).

Polyphenols

Two studies of the polyphenol constituents from *E. helioscopia* have been reported: from *E. helioscopia* collected from Uzbekistan among various *Euphorbia* species growing in the Fergan valley, quercetin, quercetin-3-O-glucoside, and 1,2,3-tri-O-galloyl-β-D-glucose (Abdulladzhanova NG *et al*., 2003) were isolated. Form Chinese species: gallic acid, methyl gallate, pyrogallol,(-)-shikimic acid-4-O-gallate, (-)-shikimic acid-O-gallate, 1-O-galloyl-2,3-HHDP-α-D-glucose, 1,3,6-tri-O-galloyl-β-D-glucose, 1,2,3,6-tetra-O-galloyl-B-D-glucose, resorcinol, gallic acide-4-O-(6`-O-galloyl)-β-D-glucose were isolated (Feng WS *et al*., 2009). The chemical constituents of fruits and roots of *E. helioscopia* were analyzed by a high- performance liquid chromatography. The major components were quercetin, quercitrin and subgallate (He XG *et al*., 1978).

Glycosides

The whole plant of *E. helioscopia* L. were collected in Xixia county, Henan province, China, extracted with 50% aqueous acetone, yield a new aryl glycoside, 3``-O-galloyl-benzyl-D-α-L-rhamnopyranosyl-(1-6)-β-D-glucopyranoside (63) (Feng WS *et al*., 2009).

Lipids, fatty acids, waxes & hydrocarbons

In a study of the neutral lipids from the leaves of *E. Helioscopia,* wax esters composed of lauric, 1.35%, myristic 5.24 %, palmitic 39.30%, stearic 13.27%, oleic 15.66%, linoleic 2.30%, arachidic 19.14%, behenic acid 3.80% and higher fatty alcohols were isolated. Octacosyl alcohol and β-sitosterol were found in both in the free and esterified form. Heptacosane and triterpenoidal acetate ($C_{32}H_{52}O_2$) were isolated from the hydrocarbon fraction, and the terpenoidal ester fraction respectively (Nazir MA *et al*., 1977). In a separate study, the neutral lipids were extracted with hexane in 2.8% yield and resolved into an acidic fraction (28.7%) and a neutral fraction (71.1%). The normal monocarboxylic acids (19.26%), the hydrocarbons (9.94%), the monohydric alcohols. (35.53%), and the sterols (5.61%) were isolated from the acidic fraction and the neutral fraction by column chromatography. In the anlysis by gas liquid chromatography saturated and unsaturated fatty acids ranging from lauric (C12) to cerotic (C26) were present with palmitic acid (C16) being the most predominant. Alkanes ranged from henocosane (C21) to heptatriacontane (C37) with hentriacontane (C31) as the major product. The alkanols ranged from behenyl (C22) to myricyl (C30) with ceryl (C26) as the max. β-Sitosterol was the major component (97.35%) of the sterol fraction (Nazir M *et al*., 1983).

The epicuticular waxes of *E. helioscopia* were fractionated into fatty acids, hydrocarbons, wax esters, aldehydes, methyl esters, triterpenol acetates, alcohols, sterols and polar components. The components of the fractions were determined by gas chromatography GC, GC-mass spectrometry, and HPLC. The main components within these lipid classes were hentriacontane the wax esters C-46 and C-48, octacosanal, hexacosanol and octacosanol, hexadecanoic acid and β-sitosterol. Lupeol and its acetate were also confirmed. (Nazir M *et al*., 1993).

The distribution of hydrocarbons in the surface wax of *E. helioscopia* was also studied. In addition to homologous series of *n*-alkanes, minor quantities of unsaturated and branched hydrocarbons were also detected. Some of the branched chain hydrocarbons could be explained as having originated from isoprene units and the substituents corresponding to diterpenes and triterpenes (Ahmed W *et al*., 1996).

E. helioscopia seeds contain 28% oil (Hossain MG *et al*., 1993), the oil isolated from the seeds of *E. helioscopia* and the natural mixture of fatty acids derived from the oil contains lauric acid 2.85 %, myristic acid 5.49 %, palmitic acid 9.18 %, stearic acid 1.13 %, oleic acid 15.80 %, linoleic acid 22.14 %, and linolenic acid 42.71 % (Durrani AA *et al*.,, 1967; Nazir M *et al*., 1986; Vioque J *et al*., 1994; Doe J, 2010; Yamamura S *et al*., 1981).

Volatile oil

Only two studies on the volatile oil of *E. helioscopia* have been reported. In Saudi Arabia, *E. helioscopia* amongst other local plants was investigated for their volatile oil constituents, with the major constituents being elemol and β-eudesmol (Baghlaf AO *et al*., 1983). The analysis of steam volatile oil obtained from the inflorescence of *E. helioscopia* was reported; resulted in the identification and quantification of 40 constituents (94.3%) were identified and quantified. The major compounds were phytol (21.2%), trans-Caryophyllene (10.0%) and docosanoic acid methyl ester (8.1%) (Fokialakis N *et al*., 2003).

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Biological Activities

Vasodepressor Activity

The crude extract of the Turkish *E. helioscopia* were partitioned against petroleum ether and then CH_2Cl_2, which give 4 fractions (A-D) thus fractions were further submitted to silica gel column chromatography to yield 7 pure compounds. The fractions and the compounds were tested for their vasodepressor effects using Wistar Albino rats. Among the compounds, 5,11-jatrophadiene-3-benzoyloxy-7,9,14-tri-acetyloxy-15-ol (45) was the most active vasodepressor (42 mmHg). The period for the effective reduction of blood pressure was 45 min. this effect lasted 70 min and did not return to normal during this period. Compound lup-20(29)-ene-3-acetate (57) dropped blood pressure by about 34 mmHg; this effect continued for 45 min. compound stigmast-4-ene-3-one (62) had the lowest vasodepressor effect (28 mmHg); however, it returned to normal after 30 min. the vasodepressor effect of these compounds might be due to vasorelaxation activity (Barla A *et al*., 2006).

Anti-Allergic & anti-asthmatic activity

A study by Park (2001) indicates an inhibitor of leukotriene D_4-induced tracheal contraction was isolated from *E. helioscopia* this isolated polyphenol compound, known as helioscopinin-A showed a certain inhibitory activity on capillary permeability in passive cutaneous anaphylaxis responses of rats and also on antigen-induced bronchial constriction in an experimental asthma model of guinea pigs. The compound at a high concentration weakly inhibited histamine release from isolated mast cells of rats. It is suggested that this compound is an anti-allergic and anti-asthmatic which exerts its activity through antagonism on leukotriene D_4-induced responses.

Allelopathic effect

Studies by Tanveer (2010) investigating the Allelopathic effect of root, stem, leaf, and fruit water extracts and infested soil of *E. helioscopia* L. on the seed germination and seedling growth of wheat, chickpea, and lentil were conducted in a completely randomized design with 4 replications. Water extracts of root, stem, leaf, and fruit were prepared by soaking dried plant parts of *E. helioscopia* in water (1:20 w/v) for a period of 24 h. Seedling emergence, seedling vigor index, and total dry weight of wheat, chickpea, and lentil seedlings were significantly reduced when these crops were grown in soil taken from an *E. helioscopia* infested field compared to soil collected from an area free of any vegetation. *E. helioscopia* infested soil also significantly decreased the root length of wheat and lentil, and shoot length of lentil compared to the control soil. Water extracts of various organs of *E. helioscopia* significantly decreased the seedling vigor index and growth of test crops. Leaf extract had a greater inhibitory effect than the other extracts. Water extracts from the root, stem, leaf, and fruit of *E. helioscopia* resulted in a reduction in the seed germination (chickpea and lentil only) and germination index but the leaf extract increased the mean germination time in all test crops.

Insulin secretagogue activity

A study by Hussain (2004) of *E. helioscopia* amongst medicinal plants, collected from Islamabad and the Murree region of Pakistan, were carried out to look into the effect of these medicinal plants on insulin secretion from INS-1 cells. INS-1 cells secrete insulin without peracrine influence dried ethanol extracts of all plants were dissolved in ethanol and DMSO, and tested at various concentrations (between 1 and 40 µg/mL) for insulin release from INS-1 cells in the presence of 5.5 mM glucose. Glibenclamide was used as a central. Promising insulin secretagogue activity in various plant extracts at 1, 10, 20 and 40 µg/mL was found, while *E. helioscopia* was active at 10 µg/mL ($p < 0.05$).

P-glycoprotein & BCRP- inhibiting activity

The isolated compounds; jatrophane diterpenes (50-52, 6, 8, 39, 43) and lathyrane (59) from the whole plant of *E. helioscopia* L. exhibited in vitro activity as inhibitors of P-glycoprotein (ABCB1) among them epieuphoscopin B (39) behaved as the most potent inhibitor of mitoxantrone efflux activity, being twice as efficient as the reference inhibitor cyclosporine A. structure activity relationships among jatrophanes showed the importance of substitution at positions 7 and 9. Interestingly, these compounds appear to be specific P-glycoprotein inhibitors since they show an absence of significant activity against BCRP (ABCG2), despite the high substrate overlapping of these transporters, thus including them in the third-generation class of specific multidrug transporter modulators. (Barile E *et al*., 2008). The jatrophane compounds and lathyrane diterpenes by Corea (2009) from the *E. helioscopia* investigated for their Pgp- and BCRP-inhibiting properties, appeared to be specific inhibitors of Pgp since they showed no significant activity against BCRP, thus resembling to the third generation class of specific MDR inhibitors. Thus, owing to the bulk availability of *Euphorbieae* plants and the relatively easy isolation of the major constituents of the diterpenoid fraction, these plants can be qualified as an interesting source of bioactive chemotypes for detailed structure-activity studies on emerging new classes of lead compounds.

Antibacterial activity

E. helioscopia amongst 109 species of Iranian plants were screened for antimicrobial activity. The results show that *E. helioscopia* was active against *Bacillus anthracis* and inactive against *Klebsiella pneumoniae*, *Proteus vulgaris*, *Shigella sonei*, *Vilorio cholerae*, *Escherichia coli*, *Saphylococcus aureus* and *Salmonella paratyphi* A. (Surmaghi SMH *et al*., 1993). The solvent extracts of *E. helioscopia*, which were extracted by using several solvents with different polarities (Kim JY *et al*., 2007), were prepared for utility as natural preservatives. The *E. helioscopia* extract by 80% ethanol was sequentially fractionated with n-hexane, dichloromethane, ethylacetate, and butanol. In order to effectively screen for a natural preservatives agent, the antimicrobial activities and cell growth inhibition were investigated for each strain with the different concentrations of *E. helioscopia* extracts. Antimicrobial activities were shown in ethylacetate fraction of *E. helioscopia*; however, ethanol, butanol and water fractions showed weak antimicrobial activity against the tested microorganisms. Among the five fractions, ethylacetate fraction showed the highest antimicrobial activities against microorganisms tested, such as *Bacillus sublitis*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella Enteritidis* and *Salmonella Typhimurium*. The polyphenol content from ethanol, n-hexane, dichloromethane, ethylacetate, butanol, and water fractions were 207.46 mg/g, 45.45 mg/g, 138.23 mg/g, 678.02 mg/g, 278.91 mg/g, and 63.76 mg/g, respectively. Antibacterial activity of the Dichloromethane and methanol extracts of the aerial parts of *E. helioscopia* was performed against *Eschericha coli*, *Bacillus subtilis*, *Shigella flexenari*, *Staphylococcus aureu*, *Pseudomonas aeruginosa* and *Salmonella typhi*. Both the extracts exhibited non-significant activity against *Bacillus subtilis* and *Salmonella typhi* at the concentration of 3 mg /ml (Uzair, 2009). In the study of the Petroleum ether, dichloromethane, methanol extracts of *E. helioscopia* were tested by Chaudhry (2001) for antibacterial activity against *Sacina leutea* and *Escherchia coli*, only the methanol extract show antibacterial activity. Meanwhile, Loothar and Choudhary (2009) stated that dichloromethane extract of the aerial parts of the plant showed non-significant activity against *Salmonella typhi* and *Bacillus subtilis*.

Antifungal activity

The fungistatis of 14 plant extracts including *E. helioscopia* against *Botrytis cinerea*, *Rhizoctonia, solani*, *Fusarium oxysporum*, *Cladosporium cucumerinum* and *Alternaria solani* was tested in vitro using growth rate and spore germination methods. The concentration of the extracts was 0.1 g/mL. The results showed that the extract from *E. helioscopia* had more than 90% inhibition rate to the spore germination of at least one fungus tested (Shunyi Y *et al*., 2006). Dichloromethane and methanol extracts of the aerial parts of *E. helioscopia* were tested against *Trichphyton longifusus, Candida albicans, Aspergillus flavus, Microsporum canis, Fusarium solani* and *Candida glabrata*. Dichloromethane extract showed 90% Inhibition against *Fusarium solani*, at the concentration of 400 μg /ml for incubation period of seven days at 27 °C with reference to miconazole as standard. While methanol extract was found to be inactive (Uzair M *et al*., 2009). Petroleum ether, dichloromethane, methanol extracts of *E. helioscopia* were tested by Chaudhry (2001) for their antifungal activity against *Claudosporium cucumerinum*, the three extracts were devoid of antifungal activity. Loothar and Choudhary (2009) stated that dichloromethane extract of the aerial parts of the plant exhibited significant activity against *Fusarium solani* with 90 % Inhibition.

Antiviral activity

Ramezani (2008) investigated *E. helioscopia* extracts for the antiviral effects using plaque reduction assay. Plant extracts were prepared using Soxhlet apparatus or by maceration in methanol after applying several enriching stages of phage CP51, phage titration was performed to determine the phage concentration in phage lysate for specifying the dilution factor of the phage to be used as negative control or the next working stages. Then IC_{50} of trifluridine, as a positive control, for phage CP51 was determined. The MIC of extracts for *Bacillus cereus* was determined as 1.25 and 0.5 mg mL^{-1} for Soxhlet and maceration extracts, respectively. To determine whether the extracts have the ability to inhibit the adsorption of virus to host cell, it was pre-incubated with phage CP51 for 30 min at 25 °C. The growth and reproduction of phage was inhibited by more than 50% at concentration of 1 and 0.25 mg mL^{-1}, respectively. In order to test the effects of extract on transcription process, *Bacillus cereus*, phage CP51 and extract were incubated together. The growth and reproduction of phage was inhibited by more than 50% at concentration of 0.75 and 0.125 mg mL^{-1} or Soxhlet and macerated extracts, respectively. These results indicated that both extracts of *E. helioscopia* have considerable antiviral activity.

Cytotoxic activity

Zhang (2004) studied the crude extract of *E. helioscopia* and the isolated compounds euphoheliosnoids A-C, the crude extract exhibited cytotoxic activity against *murine leukaemia* P388 cells, but euphoheliosnoids A-C proved to be inactive. The cytotoxicities of compounds euphoscopin A (1), euphoscopin B (2), euphoheliosnoid A (6), euphoscopin C (8), euphoscopin F (27), epieuphoscopin B (39), euphornin (43), euphornin L (52) were assayed using the HL-60 cells by MTT method, and A-549 cells by SRB method. And VP-16 (etoposide) was used as the positive control with IC_{50} values of 0.04 and 0.63 μM, respectively. Compounds (52) and (27) exhibited cytotoxicity against HL-60 with IC_{50} values of 2.7 and 9.0 μM, respectively, while the other compounds were inactive (IC_{50} > 100 μM). (Tao, 2008). All isolated compounds by Lu (2008) were evaluated for cytotoxicity against HeLa human cervical carcinoma cells and MDA-MB-231 breast tumor cells. Only two of these compounds, helioscopinolide A (3) (IC50 0.11 and 2.1 *μ*M, respectively) and euphornin (43) (IC50 3.1 and 13.4 *μ*M, respectively), were found to be cytotoxic for the HeLa and MDA-MB-231 cells. All other compounds were inactive (IC50 > 10 *μ*M) for both cell lines. Seventy-three hydrolyzable tannins and related compounds were isolated from seven *Euphorbia* plants including *E. helioscopia*. Among them, 28 compounds including nine gallotannins, eleven ellagitannins and eight related compounds were selected according to structural similarity. Cytotoxicity of them on the human tumor cell lines including A-549, SK-OV-3, SK-MEL-2, XF-498 and HCT-15 were evaluated by the SRB method in vitro. 3,4,6-Tri-O-galloyl-D-glucose was shown to exhibit the most potent cytotoxic effect (4.4 μg/mL⟨ED50⟩10.3 μg/mL) (Lee SH *et al*., 1997).

Antitumor activity

Antitumor activity of the aquatic extract the root of *E. helioscopia* L. (EWE) in Vitro were studied. Viable cells count, MTT staining and colonal formation assays of three kinds of cancer cells were used to assess the antitumor activity. Determined by viable cells count, the IC_{50} values of EWE against 7721, HeLa, MKN-45 cells were 1.26, 1.98, 1.72 mg/ml respectively (72 h). Determined MTT staining, the IC_{50} values EWE against 7721, HeLa, MKN-45 cells were 1.43, 1.67, 0.97 mg/ml. Determined by colonal formation, the inhibition rate of EWE (4 mg/ml) against 7721, HeLa, MKN-45 cells were 59.8%, 66.4%, 70.5%. The results indicated that EWE had obvious antitumor activity (Cai Y *et al*., 1999). β-sitosterol, euphornin, euphornin D, euphohelioscopin A, quercetin, gallic acid, caffeic acid, Et gallate, myricetin, and hyperoside were isolated from the ethanol ext. of *E. helioscopia* L. The antitumor activities of the isolated compounds on LA795 cells were also conducted. Gallic acid and hyperoside were reported as the antitumor constituents of *E. helioscopia* L. for the first time (Yang L *et al*., 2008).

Antioxidant activity

The solvent extracts of *E. helioscopia*, which were extracted by using several solvents with different polarities, were prepared for utility as natural preservatives. The *E. helioscopia* extract by 80% ethanol was sequentially fractionated with n-hexane, dichloromethane, ethylacetate, and butanol. In order to effectively screen for a natural preservatives agent, the antioxidant activities investigated such as DPPH radical scavenging capacity, superoxide radical scavenging capacity, and xanthine oxidase inhibitory activity of the *E. helioscopia* extracts. By the screening system, we found that ethylacetate fraction had the strongest antioxidant activity in a dose‑dependent manner. From these results, it is suggested that *E. helioscopia* could be used for the ethylacetate fraction and could be suitable for the development of a food preservative (Kim JY *et al*., 2007). Uzair (2009) studied Dichloromethane and methanol extracts of the aerial parts of *E. helioscopia* L. for their antioxidant activity. The antioxidant activity (free radical-scavenging properties) of both extracts was evaluated by thin layer chromatography (TLC) autographic assay method, using 2,2-Diphenyl-1-(2,4,6 trinitrophenyl) hydrazyl (DPPH) as spray reagent. Methanolic extract appeared as a yellow spot against purple background because of the components responsible for free radical-scavenging properties when tested at 100μg concentration, whereas dichloromethane extract did not respond to DPPH.

Cholinergic activity & Brine shrimps toxicity

The cholinergic activity of the *E. helioscopia* extracts (petroleum ether, dichloromethane, methanol) was studied by using isolated guinea-pig ileum and rabbit jejunum preparations. Guinea-pig (500-600 g) of a local breed and of either sex was used for this study. The results show that all extracts were devoid of cholinergic activity (Chaudhry BA *et al*., 2001). Also the same extracts were tested for brine shrimps toxicity, the petroleum ether and dichloromethane show brine shrimps toxicity while the methanol extract had no activity.

In vitro mushroom tyrosinase activity

Nineteen hydrolyzable tannins isolated from the *E. helioscopia* were tested for the inhibitory effect on mushroom tyrosinase activity in vitro. Inhibitory effect of gallotannin group was more potent than that of phenolcarboxylic acid and ellagitannin groups against the enzyme activity. The inhibitory activity by pentagalloyl glucose on mushroom tyrosinase was more potent (IC50, 4.9 μM) than that of kojic acid (IC_{50}, 8.7 μM) (Kim JJ *et al*., 2001).

Molluscicidal activity

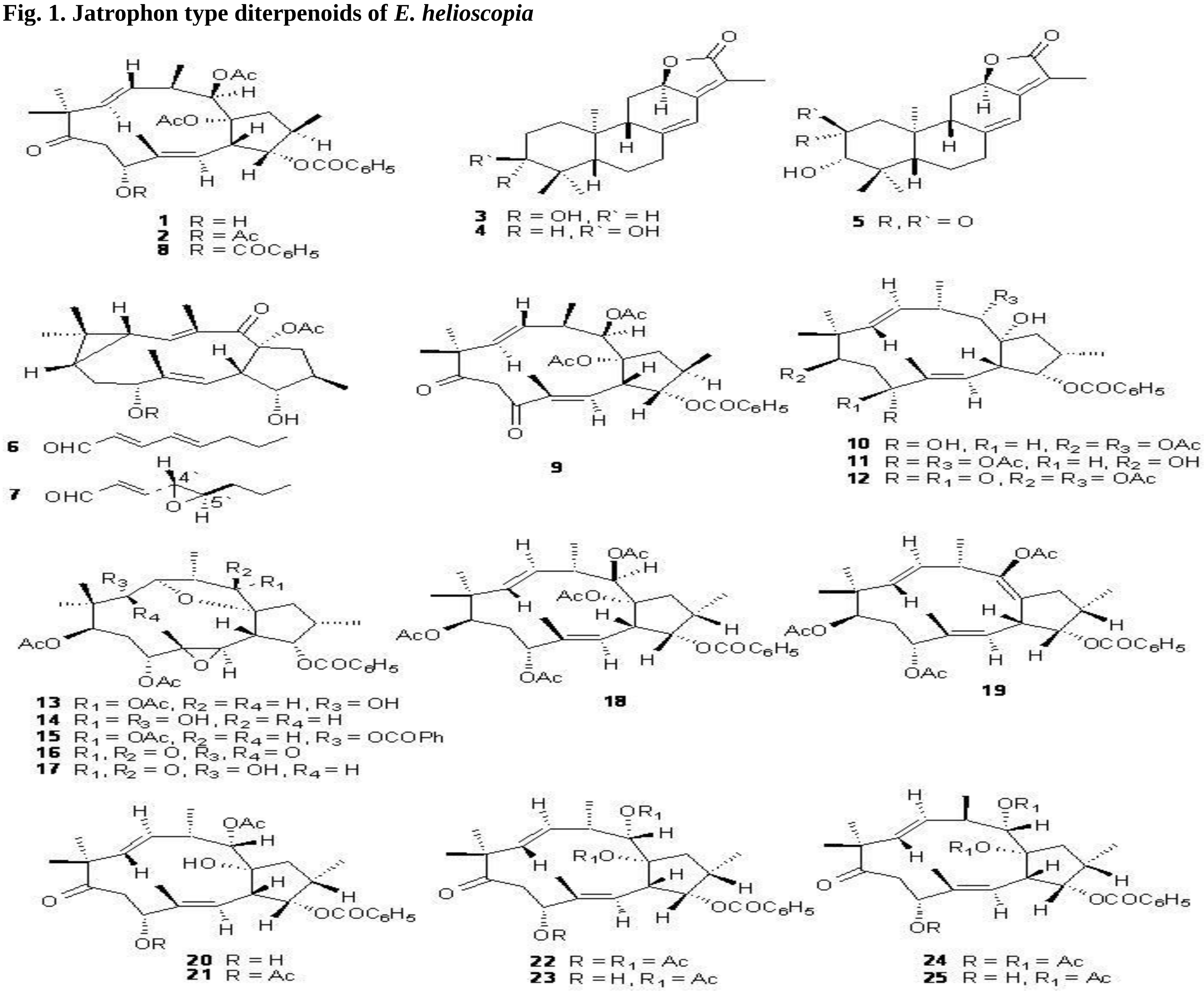
Molluscicidal activity is widespread in the family Euphorbiaceae, although activity varies greatly from species to species and even between different parts of the same plant. Al-Zanbagi (2000) studied *E. helioscopia* together with two other plants from the family Euphorbiaceae from Saudi Arabia to identify those parts of the plants that had molluscicidal activity against

Biomphalaria pfeifferi. The results showed that extracts of *E. helioscopia* and *E. schimperiana* both showed promise as molluscicides. The methanol extract of dry leaves of *E. helioscopia* had an LD_{50} of 50.8 ppm and an LD_{90} of 68.2 ppm. Using acetone extracts of the same plant Shoeb and El-Sayed (1984) and El-Amin and Osman (1991) recorded higher activities than those obtained in this study. Later, Al-Zanbagi (2005) studies the Molluscicidal properties of the *E. helioscopia* against the snail *Bulinus wrighti*. The results showed that the cold water and hot water extracts of *E. helioscopia* gave good results (LC_{50} of 80 ppm and 96.6 ppm respectively) against the snails *Bulinus wrighti*. The methanol, acetone and hexane extracts gave a big difference results against the snail *Bulinus wrighti* rather than reported for *Biomphalaria pfeifferi*. The choloroform extract has less molluscicidal activity against *Bulinus wrighti* in comparison with those of *Biomphalaria Pfeifferi*.

Pesticidal activity

Some extracts of *E. helioscopia*, *Calendula micrantha* and *Azadriachta indica* were screened for the control of *Culex pipiens* larvae, the vector of Filariasis and *Biomphalaria alexandrina* snails, the vector of Schistosomiasis in Egypt. Results showed that the acetone extract of *E. helioscopia* was the most toxic extract against both *C. pipiens* larvae and *B. alexandrina* snails with LC_{50} of 50.58 and 10.13 ppm respectively, whereas the benzene extract showed the lowest activity with LC_{50} = 98.01 and 30.1 ppm against both pests respectively. Other extracts showed moderate toxicity towards the two pests (Elyassaki WM *et al*., 1996). In a study carried out by Heng-Guo H.E. (2010), ethanol extract from *E. helioscopia* showed contact toxicity (LC50= 870.25 mg/L) and antifeedant activity (AF50 of antifeeding ratio was 23.71 mg/L.) against 2nd instar larvae of the Lepidopteron moth, *Pieris rapae*.

Fig. 1. Jatrophon type diterpenoids of *E. helioscopia*



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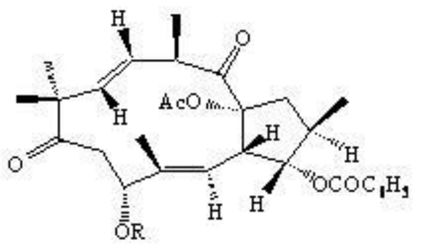
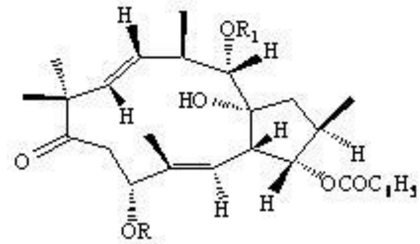
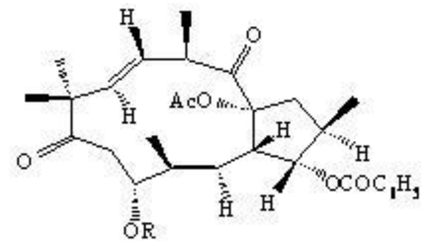


26 R = H
27 R = Ac



28 R = H, R_1 = Ac
29 R = R_1 = Ac
30 R = Ac, R_1 = H



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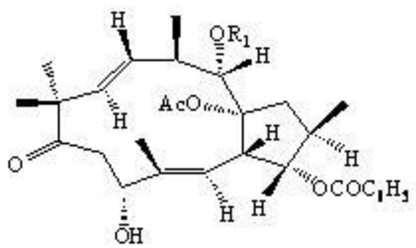
32 R = Ac



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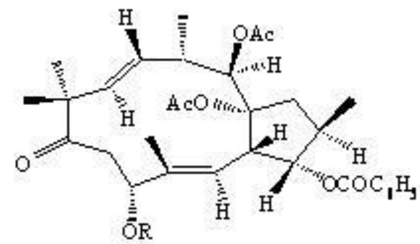

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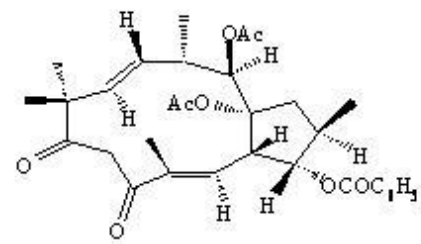
35 R = Ac



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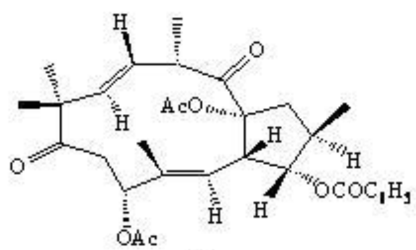

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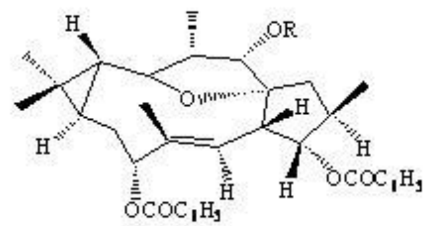

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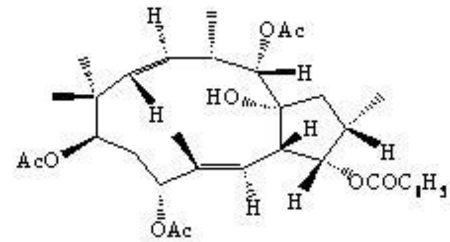

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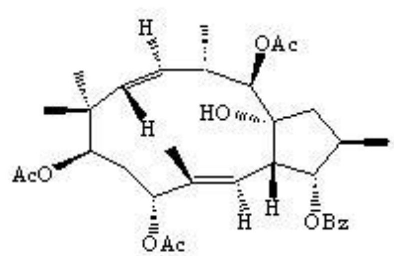

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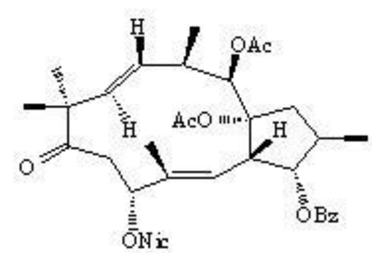

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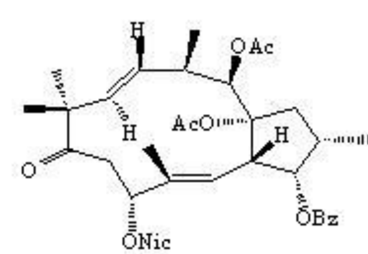

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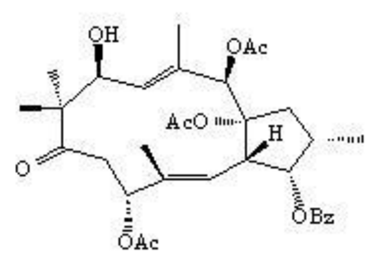

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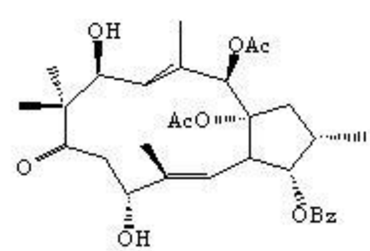

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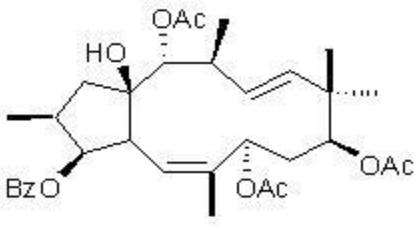

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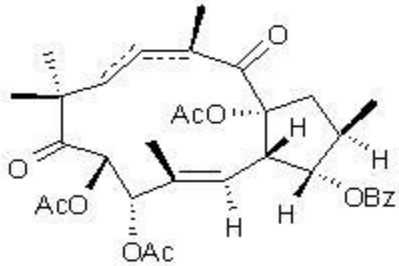
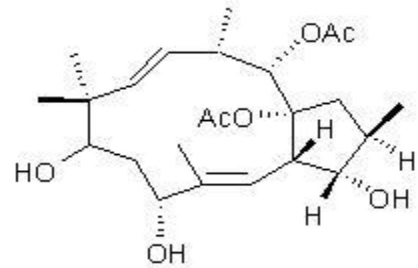


46 $\Delta^{11,12}$
47 $\Delta^{12,13}$



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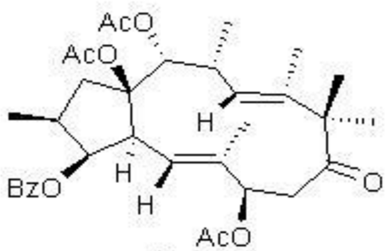

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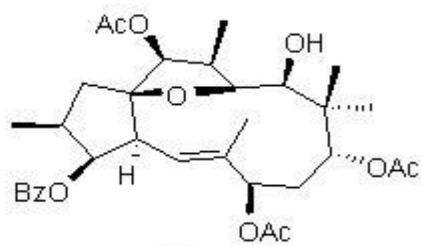

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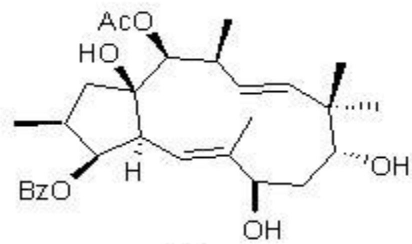

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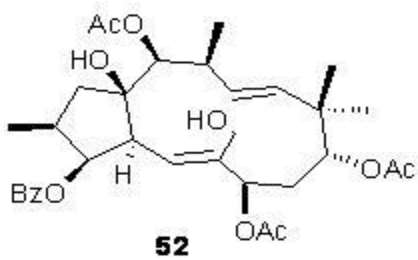

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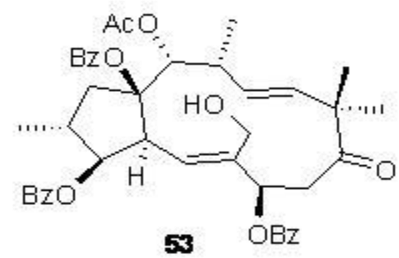

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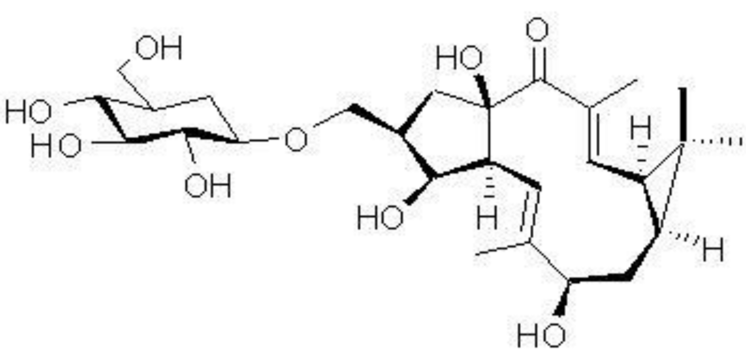
Fig. 2. Lathyrane diterpenes of *E. helioscopia*



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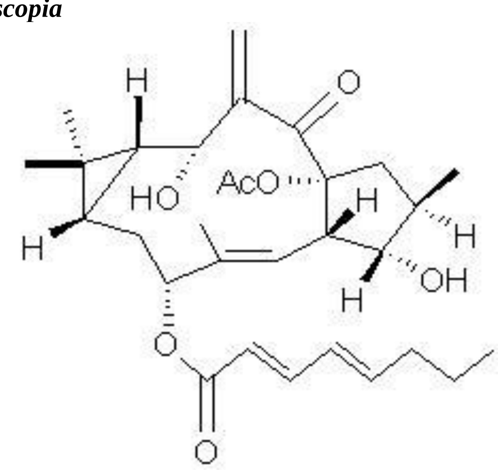

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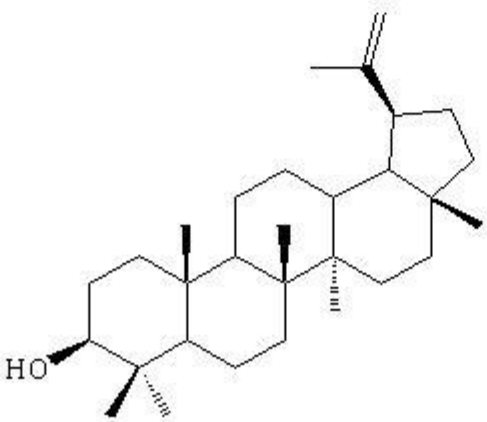
Fig. 3 Triterpenes of *E. helioscopia*



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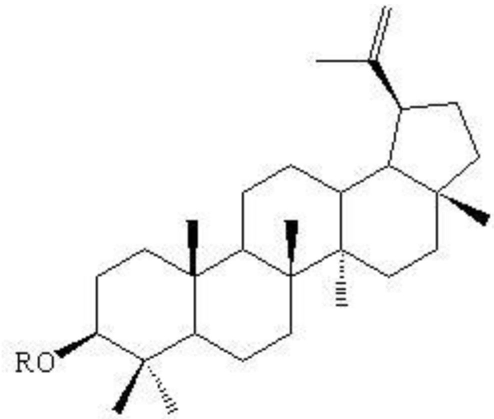
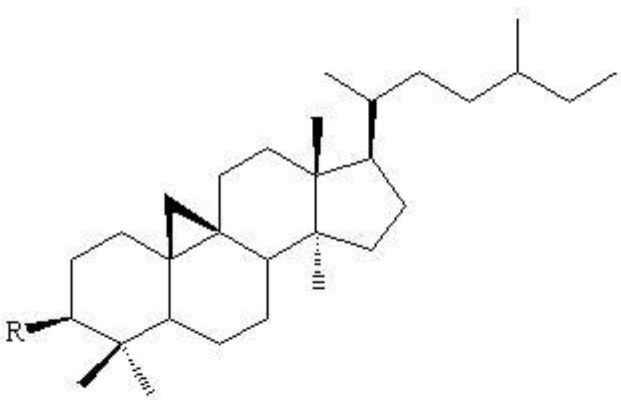


57 R = $CH_3(CH_2)_{14}CO$
58 R CH_3CO



59 R = β-OH, α-H, R_1 = H
60 R = O, R_1 = CH_2
61 R = β-OH, α-H, R_1 = CH_2

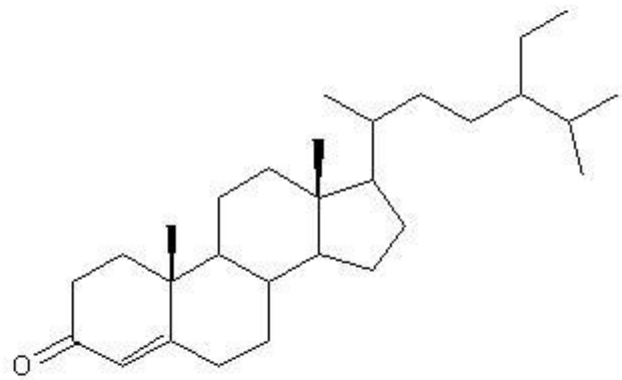


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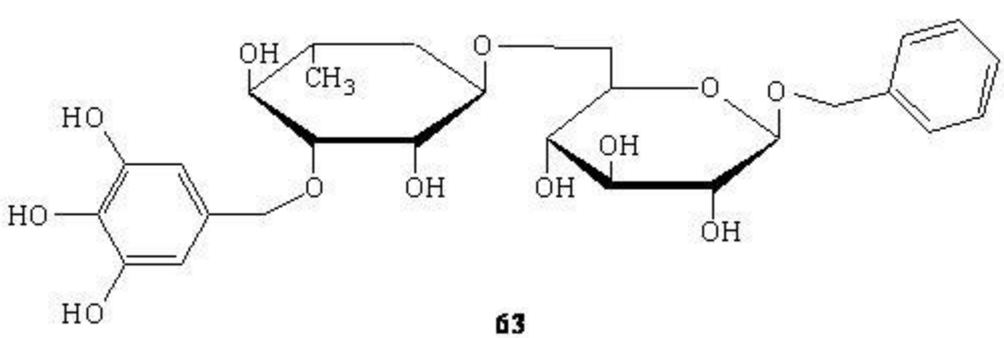
Fig.4 Glycosides of *E. helioscopia*



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Conclusions and future perspectives

The available literature indicating various biological and therapeutic activities of *E. helioscopia*. Large scale experiments would be required to substantiate the efficacy of the different classes of secondary metabolites isolated from this plant. For the wide scale and commercial use of this plant, trials should be done to validate the relevant concentrations and the economic values of using these biorationals in different biological and therapeutic applications. Assessments have to be extended to establish various limitations about their mammalian and environmental safety.

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