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# **ORIGINAL ARTICLE**

# An Environmentally-Friendly Method for Exploitation of Oleo-Gum-Resin from

# Pistacia atlantica Desf. Trees

Maryam Didehvar, Mohammad-Taghi Ebadi<sup>\*</sup>, Mahdi Ayyari Nooshabadi

Department of Horticultural Sciences, Tarbiat Modares University, Tehran, Iran

# A R T I C L E I N F O A B S T R A C T

Keywords: Essential oil; Ethrel; Gum; *Pistacia atlantica* Desf; Saqez; α-pinene Pistacia atlantica Desf.is one of the main wild species of Pistacia in Iran and one of its products is an oleo-gum-resin briefly called Saqez gum. The effect of Ethrel injection (EI) on Saqez gum production was studied during summer 2018 at three habitats (Kuh-e Birk, Goharkuh and Mirabad) inSistan and Baluchestan province, Iran. Four treatments, 0 (distilled water), 10, 20 and 30% concentration of Ethrel, were administered by injection into the tree trunk. The investigation was carried out in a randomized complete block design and 36 trees were injected. Saqez gum essential oil (EO) was extracted by Clevenger apparatus and identification and quantification of EO components were performed by GC-MS and GC-FID, respectively. The results showed that applying the EI treatment increased the gum yield of trees by an average of 4.4 times. The highest gum yield (29.33 g/tree) was obtained with EI at 10% in the Kuh-e Birk habitat, while the lowest yield (3.33 g/tree) was observed in the control treatment of the Mirabad habitat. In total, 39 compounds were identified in the EOs and  $\alpha$ -pinene was found to be the predominant compound in all samples (66 to 91.8%). Other compounds such as  $\beta$ -pinene, camphene, sabinene, *cis*-limonene oxide, neo-iso-verbanol,  $\rho$ -cymene and  $\rho$ -cymenene were identified in the EOs. EI treatments did not have a significant effect on EO components and the amount of  $\alpha$ -pinene and  $\beta$ -pinene in EI 30% treatment increased slightly. Due to the desired quantity and quality of gum in the EI 10%, this method of exploitation can replace the traditional method.

### Introduction

Trees and shrubs have many benefits for humanity and in addition to their importance in the environment and the production of food products, some industrial materials are also exploited by them (Hassanzadeh *et al.*, 2017; Yadegari *et al.*, 2018; Guerra and Scremin-Dias, 2018; De Souza *et al.*, 2021). The genus *Pistacia* has 12 species in the world, among which there are three species found in Iran (*P. vera*, *P. atlantica*, and *P. khinjuk*). *P. atlantica* Desf., commonly known as the Mt. Atlas mastic tree or Persian turpentine tree, or Baneh in the Persian language, is mainly distributed in the western parts and to a lesser extent in the eastern and central parts of Iran, covering an area of 1,200,000 hectares. The Mt. Atlas mastic tree is dioecious, with a height of 2 to 7 meters, the leaves are alternate, compound and the flowers are pink. Flowering occurs before vegetative growth, and the fruit type is drupe in various shapes and sizes (Gourine *et al.*, 2013;

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Mozaffarian, 2013; Yousefi et al., 2020; Behzadi et al., 2021).

One of the most important products of P. atlantica is an oleo-gum-resin called turpentine gum or Saqez. In the pistachio family, resin ducts are naturally produced in young seedlings and they are present in trunk and shoots, leaves, and flowers. The best season for harvesting Saqez gum is the summer, and generally, when the weather is warm, a warm climate causes more oleo-gum-resin exudation (Mahdavi, has many 2011). Sagez gum nutritional, pharmaceutical, and industrial properties (gum, pharmaceutical, paint, polymer, wax, and leather

industry). It also has antiseptic, laxative, and soothing properties (Sadeghi *et al.*, 2016). Oleo gum resins play an important and vital role in the economy of rural communities, and it is essential to find methods to harvest them sustainably without destroying trees (Ballal *et al.*, 2005; Ashmawy *et al.*, 2018). Of course, increasing their yield and quality is also important. The traditional method of gum harvesting is associated with severe damage to the tree trunk by axes and trees are more likely to die from these injuries (especially in arid areas where wound healing is difficult) (Fig. 1).



Fig. 1. Common methods of *P. atlantica* gum exploitation.

An easy and safe method of increasing oleo-gumresin exudation has been developed using ethephon to improve gum yield and wound healing (Nair, 2004). Ethrel is a plant growth regulator of the ethylene group with the common name Ethephon. Ethephon (2chloroethyl phosphonic acid) is a synthetic form of ethylene, which is commonly used as a plant growth regulator. Ethephon is a safe, low-cost, and non-toxic compound. It is used for increasing rubber yield in Ficus elastica, ripening of climacteric fruits (banana, persimmon, avocado, etc.), stimulation of abscission, flowering, root initiation, and seed germination, and breaking of dormancy (Nair, 2000). Ethephon is a stable and liquid compound under low pH, but with a rise in pH it is unstable. Plant cells absorb this compound in liquid form and neutral pH of cytoplasm, ethephon degrades and produces ethylene, which triggers the production of endogenous ethylene and leads to flower and fruit abscission. Ethylene

assimilate translocation which increases the water potential of the xylem, and assimilate restriction results in fruit abscission because of this competition (Afshari-Jafarbigloo *et al.*, 2020). Ethylene is usually also biosynthesized in plants in reaction to abiotic stresses, especially water deficit (Kanzaria *et al.*, 2015; Raj and Sing, 2017; Nazoori *et al.*, 2022). The use of ethephon improved the exudation of oleo-gum-resin in several hardwood species (Nair, 2000). Harsh *et al.* (2013) investigated thirty Gum

induces callus synthesis, which blocks the vascular

system, this leads to a reduction in water and

2000). Harsh *et al.* (2013) investigated thirty Gum Arabic trees of similar age, size, and vigor with various doses of ethephon in a season and stated that the trees started exuding gum tears 8–10 days after the EI. Average gum yields were 39.9, 67.7, and 140.6 g per tree, under EI at 9.75, 14.62, and 19.5% concentrations, respectively. They also recommend March to June as the best period for the employment

of this technique in India. Nair (2004) concluded that after ethephon application in Karaya trees, the yield was improved about 20 to 30 times more than the control and about 10 times more than when using a traditional technique. In a study, the gum-inducing chemical and ethephon were injected into the stem of Acacia Senegal trees. The gum inducing chemical and ethephon resulted in 244.37% and 200.37% more gum production over the control, respectively (Kanzaria et al., 2015). Babu and Menon (1989) observed that ethephon application in the two tree species had different results. They used 0, 240, 480 or 720 mg ethephon for Bombax ceiba and 0, 96, 192, 480, 768 960 ethephon for Sterculia urens or mg (Concentrations used per 2 ml water). In B. ceiba, the highest yields belonged to 240 mg (282 g gum) and 480 mg (378 g) treatments and there was no exudation from the 0 mg treatment. In S. urens, the highest yield was obtained from the 768 mg treatment (64.5 g), followed by the 960 mg treatment (38 g), while an insignificant level of exudation (0.5 g) resulted from the 0 mg treatment. In another study, 40 and 120 mg ethephon applications were compared to controls during field research in a semi-arid and a sub-humid region in Northern Cameroon. The gum yield was increased by 400-600% when compared to untreated trees. Gum yield at the semi-arid location was 77, 313, and 214 g/tree with 0, 40, and 120 mg ethephon/tree, respectively, while at the sub-humid location, it was 30, 186, and 114 g/tree with 0, 40, and 120 mg ethephon/tree (Abib *et al.*, 2013).

The traditional method of gum harvesting causes the death of many trees, especially in arid areas, and new methods need to be employed which result in minimal damage to trees (Gaafar *et al.*, 2006; Saini *et al.*, 2018). No research has been done on novel methods of exploiting Saqez gum and the effect of EI on increasing the oleo-gum-resin content of *P*. *atlantica* was unknown until now. Therefore, this investigation aimed to evaluate the effect of EI on Saqez gum secretion, its compounds, and the survival rate of treated *P. atlantica* trees in later years.

## **Materials and Methods**

### Plant material

This study was conducted in the summer of 2018 in three *P. atlantica* habitats in Sistan and Baluchestan province, Iran (Fig. 2). In this study, trees with the same trunk diameter, height, and vigor were selected. Geographical coordinates and collection site information for the three *P. atlantica* populations are shown in Table 1.

No.	Habitat	Longitude	Latitude	Altitude (m.a.s.l)	Average annual temperature (C°)	Average annual rainfall (mm)		
1	Kuh-e Birk	62 8 33.046	27 50 19.035	1594	22.2	112.4		
2	Gohar Kuh	60' 31' 7.385"	28° 23 <sup>°</sup> 20.706 <sup>°</sup>	1384	20.2	142.8		
3	Mirabad	61 16 50.299	28 16 45.032	1869	20.2	142.8		

Table 1. Geographical coordinates and collection sites Information for three P. atlantica populations in Sistan and Baluchestan province, Iran.



Fig. 2. Map of collection sites for three P. atlantica populations

## Ethrel injection (EI)

The research was carried out in randomized complete block design (RCBD) with four treatments and three replications. Ethrel (Ethrel® 480 SL, Bayer, Germany - Containing 480 g.a.i./l Ethephon (2-chloroethyl phosphonic acid)) was used to induce gummosis. The treatments consisted of injection of  $T_1$  (Distilled water = control),  $T_2$  (Ethrel 10%),  $T_3$  (Ethrel 20%), and  $T_4$  (Ethrel 30%) that they prepared using distilled water, volumetrically (v/v). In treatments, a 5 mm aqueous solution of Ethrel was injected. A hole of

1 cm in diameter and about 5 cm deep was made in the tree trunk 0.5–1.0 m above the soil, using a hand drill. This method was used to avoid wastage of Ethrel due to outfall and evaporation. One 5-ml dose of the chosen concentration of Ethrel was injected into the hole. Immediately after the injection, the hole was covered with moistened clay. The whole process is shown in Fig. 3. Two weeks after the treatment, Saqez gum was collected for the first time.



Fig. 3. Ethephon-induced gum exudation technique. (A) Making hole with a hand drill, (B) Injecting ethephon solution, (C) Covering whole with moistened clay, (D) Exudated oleo-gum-resin

### Essential oil isolation

To determine the amount of EO, 2 g of Saqez gum was hydro-distilled in three replications by Clevenger apparatus for 3 hours, and the EO was calculated (v/w %). The EO was collected, dried over anhydrous sodium sulfate, and stored at 4°C until analysis (Demirci *et al.*, 2001).

# Gas Chromatography (GC) and Gas Chromatography-Mass Spectrometry (GC-MS)

The EOs were evaluated by a GC (Agilant 7890B) equipped with a flame ionization detector, and an HP-5 capillary column (length 30 m, internal diameter 0.25 mm, and 0.25 µm film thickness). The temperature program includes 2 min at 60°C, increased to 280°C with a ramp of 5°C/min. GC-mass spectrometry (GC-MS) analysis was done using a Thermoquest-Finnigan gas chromatograph equipped with a fused silica capillary HP-5 column (60 m  $\times$ 0.25 mmi.d.; film thickness 0.25 µm) coupled with a trace mass spectrometer. Helium gas was used as the carrier at a flow rate of 1.1 mL/minute in a split ratio of 1:150. The ionization voltage was 70 eV. Ion source and interface temperatures were 200 and 280°C, respectively. Mass range was adjusted from 45 to 456 amu. The same GC oven temperature program was used. Identification of EO constituents was confirmed by comparison of each component's mass spectra with those of the internal mass spectra library of the main library, Wiley 7.0 and Adams while further identification was based a comparison of peak

retention indices by using a homologous series of Normal Alkanes (C8 to C24) verified under the same operating situations and data published in the literature (Adams 2001).

# Statistical analysis

The investigation was done based on a randomized complete block design with three replications, using SAS Statistical Package Program version 9.0. Means compared by Duncan's Multiple Range Test at the level of probability of 0.01.

#### Results

## Gum yield

The results showed that EI had a significant effect on gum yield in all three habitats. As can be seen in Figure 4, the highest gum yield (29.33 g/tree) was related to the EI 10% treatment in the Kuh-e Birk habitat, while the lowest yield (3.33 g/tree) was related to the control treatment in the Mirabad habitat. The results of this investigation showed that *P. atlantica* trees in the Sistan and Baluchestan province, when compared to the Ilam (17.9 - 555.4 g/tree) and Chaharmahal and Bakhtiari (571.9 g/tree) provinces resulted in less gum production (Iranmanesh *et al.*, 2019; Karamshahi *et al.*,2005), which is probably due to less rainfall in this province (Das *et al.*, 2014).



Fig. 4. Gum yield of P. atlantica trees treated with ethephon

### Essential oil content

As be seen in Fig. 5, the highest amount of EO (25%) was related to the EI 10% treatment in the Kuh-e Birk habitat and the lowest amount (15%) was related to the control treatment in the Mirabad habitat,

and EI 10 and 30% treatments in Goharkuh habitat. In the studies that were done previously, no research was carried out concerning the EO content in the guminducing trees treated with Ethrel.



Fig. 5. EO content in the gum of P. atlantica trees treated with ethephon

#### Essential oil composition

In total, 39 compounds were identified in the EOs, which accounted for about 94.5% to 99.0% of the total EO.  $\alpha$ -pinene was present in all samples and the highest value was related to this compound. The lowest amount of  $\alpha$ -pinene (66%) was observed in the EO of the control treatment in the Gohar kuh habitat, while the highest amount (91.8%) was obtained using the EI 30% treatment in the Kuh-e Birk habitat (Fig. 6). Other predominant compounds such as  $\beta$ -pinene, camphene, sabinene, *cis*-limonene oxide, neo-isoverbanol,  $\rho$ -cymene and  $\rho$ -cymenene were identified. EO compounds of samples can be classified into three chemical groups: monoterpene hydrocarbons (representing 86.1–95.9% of all compounds), oxygenated monoterpenes (2.6–9.6%), sesquiterpene hydrocarbons (0.0–1.5%) (Table 2).

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				Kuh-e Birk						Goha	ırkuh			Mirabad				
NO	$\mathbf{RT}^*$	Compounds	RI <sup>cal**</sup>	Control	10%	20%	30%	C	Control	10%	20%	30%	Control	10%	20%	30%		
1	7.7	α-Pinene	936	86	88	85.2	91.8		66	70.1	70	73.3	81.2	83.4	82.7	79		
2	7.9	Camphene	954	0.9	0.7	0.8	0.6		1.3	tr***	1.1	1.1	1.2	0.5	0.7	1.1		
3	8	Thuja-2.4(10)-diene	958	0.1	0.1	0.1	0.1		0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1		
4	8.4	Sabinene	976	0.3	0.3	0.3	0.3		9.3	4.5	5.7	1.8	0.1	0.9	0.4	0.5		
5	8.5	β-Pinene	983	1.9	1.8	1.7	2		9.9	9.1	10.5	9.2	7.2	3.3	7.5	10.7		
6	8.7	Myrcene	991	tr	tr	tr	tr		0.1	tr	tr	tr	tr	tr	0.1	tr		
7	8.8	δ-2-Carene	995	0.1	0.2	0.1	0.1		0.1	0.1	0.1	0.1	0.2	0.1	0.2	0.1		
8	9.3	$\alpha$ -Phellandrene	1010	tr	0.3	0.1	0.1		0.2	0.1	0.3	0.1	0.5	0.7	0.7	0.7		
9	9.6	δ-3-Carene	1014	tr	tr	tr	tr		0.1	tr	tr	tr	tr	tr	tr	0.1		
10	9.7	α-Terpinene	1021	0.1	0.1	0.1	0.1		0.8	0.7	0.8	0.6	0.2	0.2	0.2	0.3		
11	9.8	ρ-Cymene	1029	0.4	0.6	0.6	0.4		1.2	1	1.2	1.1	1.1	0.7	1.1	1.4		
12	9.9	Limonene	1033	0.2	0.1	0.2	0.2		0.2	0.1	0.2	0.1	0.1	0.1	0.1	0.1		
13	10	1.8-Cineole	1037	tr	tr	0.1	0.1		0.1	tr	tr	tr	0.1	0.1	0.1	0.1		
14	10.1	<i>trans-</i> $\beta$ -Ocimene	1047	tr	tr	tr	tr		0.1	tr	tr	tr	tr	tr	tr	tr		
15	11.3	dehydro-Linalool	1091	tr	0.1	0.1	0.1		0.1	0.2	0.1	0.1	0.1	0.1	tr	tr		
16	11.5	$\rho$ -Cymenene	1096	0.2	0.5	0.3	0.2		0.3	0.3	0.3	0.2	0.9	1.2	1.6	1.3		
17	11.7	Linalool	1103	0.5	0.6	0.5	0.4		0.4	0.8	0.3	0.5	0.3	0.3	0.2	0.3		
18	12	$\alpha$ -Pinene oxide	1109	0.4	0.4	0.3	0.2		0.3	0.5	0.1	0.2	0.2	0.1	0.1	0.1		
19	13.1	cis-Limonene oxide	1134	2.9	1.8	2	1.1		2.5	3.8	2.4	2	1.7	1.8	1.2	1.1		
20	13.1	trans-Pinocarveol	1149	0.1	0.1	0.1	-		0.1	0.1	0.1	0.1	0.1	-	-	-		
21	13.3	trans-Verbenol	1153	0.1	tr	tr	tr		0.1	0.1	0.1	0.1	tr	0.1	tr	tr		
22	13.6	cis-Isocitral	1166	0.1	0.1	0.1	tr		0.3	-	0.1	0.2	tr	0.1	tr	tr		
23	13.9	Pinocampheol	1170	0.1	0.1	0.1	tr		0.5	0.5	0.2	0.2	0.1	0.1	0.1	0.1		
24	14.3	trans-Isocitral	1180	0.4	0.2	0.2	0.1		0.5	0.5	0.5	0.4	0.3	0.3	0.2	0.3		

Table 2. EO compounds in the gum of P. atlantica trees were treated with different concentrations of ethephon in three selected habitats.

25	14.4	neo-iso-Verbanol	1187	0.5	0.4	0.3	0.2	0.8	1.1	0.8	0.6	0.3	0.2	0.2	0.3
26	14.8	Myrtenal	1206	0.4	0.3	0.3	0.1	0.3	0.5	0.4	0.3	0.3	0.3	0.1	0.2
27	15	Verbenone	1218	0.2	0.1	0.1	tr	0.1	0.3	0.2	0.1	0.1	0.1	tr	0.1
27	15.1	cis-Carveol	1227	-	0.1	tr	-	0.1	-	-	-	0.1	0.1	tr	tr
29	17	$\rho$ -Menth-1-en-7-al	1283	0.1	0.1	0.1	tr	0.1	0.1	0.1	0.1	tr	tr	tr	tr
30	17.2	Bornyl acetate	1289	0.9	0.6	0.7	0.3	0.3	0.6	0.5	0.6	0.4	0.3	0.2	0.4
31	17.4	Carquejol acetate	1300	0.2	0.2	0.1	0.1	0.1	0.2	0.2	0.2	0.1	0.2	0.1	0.2
32	17.5	Carvacrol	1307	0.5	0.3	0.4	0.1	0.2	0.3	0.3	0.4	0.2	0.1	0.1	0.2
33	18.5	α-Cubebene	1349	tr	tr	0.2	tr	tr	tr	0.1	tr	tr	tr	-	tr
34	18.5	α-Longipinene	1354	tr	-	tr	-	tr							
35	19.2	α-Copaene	1379	-	tr	0.2	tr	tr	tr	tr	-	tr	0.7	-	tr
36	20.3	trans-Caryophyllene	1410	tr	tr	tr	-	tr	tr	tr	tr	tr	0.1	0.1	0.1
37	21.9	β-Selinene	1495	tr	tr	0.2	-	-	tr	-	0.3	0.3	0.7	tr	tr
38	22.7	δ-Cadinene	1522	tr	tr	0.1	-	tr	tr	tr	0.4	tr	-	-	0.1
39	23.2	cis-Calamenene	1528	-	tr	tr	tr	tr	tr	-	tr	-	-	-	-
Monoterpene hydrocarbons			90.2	92.7	89.5	95.9	89.7	86.1	90.3	87.7	92.8	91.2	95.4	95.4	
	Oxygenated monoterpenes			7.4	5.5	5.5	2.8	6.9	9.6	6.4	6.1	4.4	4.3	2.6	3.4
	Sesquiterpene hydrocarbons				0	0.7	0	0	0	0.1	0.7	0.3	1.5	0.1	0.2
Total				97.6	98.2	95.7	98.7	96.6	95.7	96.8	94.5	97.5	97	98.1	99

\*: RT: Retention time \*\*: RI<sup>cal</sup>: Calculated retention index \*\*\*\*: tr: trace



Fig. 6.  $\alpha$ -pinene content in the EO of gums.

## Survival of trees

The following year (2019), the habitats were visited and no withered trees were observed; nor were their growth reduced. Using traditional methods, gum harvest is achieved by inflicting severe damage to tree trunks, which is very dangerous and threatening for young trees. Even vigorous trees can be seriously damaged and die if harvested consecutively for several years consecutively (Bhatt *et al.*, 1989). Therefore, the method examined in this research can be introduced as an environmentally-friendly method.

#### Discussion

The results indicated that employment of the EI treatment increased gum yield by 0.3 to 4.4 times. Similar results have been reported in stimulating gum and resin production in trees through the use of ethephon. For example in the Gum Arabic tree, Harsh et al. (2013) investigated the effect of EI on gum yield and the results showed that this method increased gum yield from 3.6 to 425 g per tree. In another report, Garasiya et al. (2013) stated that the maximum gum yield was recorded in 900 ppm EI treatment with an average of 386 g/tree, and the lowest gum yield was measured in the control (80.33 g/tree). Abib et al. (2013) concluded that gum yield in EI-treated trees was improved by 400-600% when compared to the untreated ones. Other researches on trees such as Prunus cerasus (Olien and Bukovac 1982), Anogeissus latifolia (Bhatt, 1987), Bombax ceiba and Sterculia urens (Babu and Menon, 1989), Commiphora wightii (Bhatt et al., 1989), Prunus domestica (Saniewskiet al., 2002), Sterculia urens (Nair, 2004), Acacia nilotica (Das et al., 2014; Raj and Sing, 2017), Anogeissus latifolia (Kuruwanshi and Pratibha, 2017) and Acacia seyal var. seyal (Ahmed et al., 2019) have reported the positive effects of EI in increasing gum and resin yield. One way to increase the yield of plant gum exudates is to use plant growth regulators. The most satisfactory results are obtained by the use of ethephon which releases ethylene. Ethylene is the compound that plants produce when exposed to environmental stresses and which makes them resistant to stresses (Abib et al., 2013). In the exploitation methods applied for P. atlantica forests, traditional methods are commonly used, which are not suitable for two reasons: Firstly, due to the extent of the wound created in a place that is not easily repaired, and secondly, due to the low production of gum in the use of these methods (Bordbar et al., 2006). making cuts on a tree trunk or peeling it to extract gum is not the right and practical method and can lead to stunted growth and even the death of trees.

Increased ethylene biosynthesis has been observed at different stages of plant growth under environmental stresses. Oleo-gum-resin exudation may be considered as one such progressive response to drought stress. The initial idea of using ethephon as a stimulant for gum secretion came from the fact that the external application of ethephon accelerated the plants' response to drought stress and produced more gum (Harsh *et al.*, 2013). Gummosis is generally observed in the plant kingdom. Gum creation in plants is promoted by environmental stressors such as infection, insect attack, flooding, and mechanical or chemical damage. All of the reasons which excite gum exudation also stimulate ethylene synthesis in plants. Ethylene or ethylene-releasing compounds such as ethephon encourage gum production in some species of the Rosaceae, Fabaceae, and Anacardiaceae families. According to previous research, ethylene is the key factor responsible for gum secretion in trees (Saniewski *et al.*, 2006).

This research was examined for the first time. However, some researchers have evaluated the EO amount of Saqez gum without examining the type of harvesting method applied. For example, in Iran, the EO content of Saqez gum from habitats in Kurdistan (20-22%), Kermanshah, Lorestan, and Ilam (18.3-23.3%) and Fars (17.2%) provinces and also in Algeria (47%) and in Morocco (32.6%) was reported (Moraghebi et al., 2001; Delazar et al., 2003; Barrero et al., 2005; Mecherara-Idjeri et al., 2008; Sharifi and Hazell, 2011; Panahi et al., 2017). The findings of this research showed that EI treatment could increase the EO amount of Saqez gum by 66% compared to the control and also caused the EO amount to be higher than samples from other parts of Iran but less than some African countries, probably due to differences in climatic conditions and genotype of trees.

 $\alpha$ -pinene is one of the main constituents of EO in conifer resin, *Pistacia* gum, rosemary, and lavender. This compound has particular economic importance and has been used in the perfume industry for many years. A considerable amount of research has been done on the use of this compound in the formulation of natural herbicides, fungicides, and insecticides (Abrahim *et al.*, 2003; Silva *et al.*, 2012). As can be seen in Table 2, EI treatments did not have a significant effect on the main components of EO and the amount of  $\alpha$ -pinene and  $\beta$ -pinene in EI 30% treatment increased slightly. α-pinene has been considered the most important component of Saqez gum EO in several studies. For example, in Iran, Moraghebi et al. (2001) reported that the amount of this compound in the EO fluctuated between 84 and 91% in Kermanshah, Lorestan, and Ilam provinces. Panahi et al. (2017) measured its amount in natural habitats of the Eghlid region of Fars province at 77.1% and Sharifi and Hazell (2011) reported it at 22% in Kurdistan province. Also, Delazar et al. (2003) stated that its amount was 70% in the Marivan region of Kurdistan province. In African countries, such as Morocco and Algeria, the amount of this compound in EO was reported to be 42.9%, respectively (Barrero et al., 2005; Mecherara-Idjeri et al., 2008). According to the results, it can be concluded that Saqez gum EO in the natural habitats of Sistan and Baluchestan province has relatively high amounts of a-pinene compared to many other regions in the world.

## Conclusions

The new method of exploiting *P. atlantica* gum by using EI led to an increase in gum exudation of trees without harming their growth and development, so that the highest gum yield was observed for trees treated with Ethrel and the lowest yield was observed in control trees. The EO composition of the three populations coming from different habitats showed the predominance of the hydrocarbon monoterpene groups, among which  $\alpha$ -pinene was the main component.

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