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Evaluation of environmental pollutions on *Pinus eldarica* needles essential oil

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Abstract

Influence of environmental stress factors on both crop and wild plants of nutritional value is an important research topic. The past research has focused on rising temperatures, drought, soil salinity, but the potential effects of increased environmental contamination by human-generated pollution on plants have little been studied. Here it studied the influence of environment pollutions on essential oil analyses in *Pinus eldarica*. The needles of *Pinus eldarica* Medw. were collected from different regions of Isfahan in Iran (1: Around of industrial factories. 2: In city center with air pollution. 3: Around of high voltage pylons) and they were analyzed by using GC/MS. The 40, 15, 19 compounds were identified in industrial pollution, air pollution and electromagnetic pollution, respectively. The major components in needles of industrial region were: Octadecenoic acid (31.52%), α-pinene (14.95%), 3-carene (4.23%), β-pinene (3.151%). The major components in in needles of collected with air pollution were: α-pinene (52.57%), β-pinene (12.21%), Octadecenoic acid (7.19%), limonene (7.07%). The major components in needles of collected from electromagnetic region were: α-pinene (29.46%), β-pinene (12.32%), limonene (4.93%), Octadecenoic acid (4.92%). Air pollution resulted in density of a-pinene, so that density of a-pinene in urban region *Pinus eldarica* needles was highest. There was a direct relationship between environmental pollutions and essential oil chemical modifications of the three plants studied. These data collectively demonstrate that human-generated pollution can potentially constitute a stress to the plants.

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Introduction

Environment pollutions are a serious problems in urban and industrial areas. Heavy metal deposition near industrial factories may affect vegetation growth and may lead to the damage and alteration of animal, including human, physiological functions through the food chains (Nouri *et al.*, 2009; Fowler *et al.*, 2009).

Heavy metals accumulation in plants from human sources has drawn greater attention to inorganic pollution, and established plants as passive biomonitors, since plants respond directly to the state of soil and air (Fowler *et al.*, 2009). Higher plants which are most often used for biomonitoring in industrial and urban areas, are coniferous and deciduous trees (Piczak *et al.*, 2003). The results of many researches demonstrate that metal accumulation in plants varies, depending on the species, tissues and metals (Serbula *et al.*, 2012).

Pine has needles with a thick epicuticular wax layer which made pine species the best suited biomonitors, particularly sensitive to environmental pollution (Mingorance *et al.*, 2007). The morphology and epicuticular wax on the leaf surface make linden trees suitable for biomonitoring of air pollution (Braun *et al.*, 2007). Pine needles are most frequently used for biomonitoring of airborne pollution, because of the possibility of passive and active uptake of needles tissue from the atmosphere (Kuang *et al.*, 2011).

The most frequently identified elements which cause biological/ecological harmful effects are Pb, Cs, As, Cr, Zn, Ni, and Cu. Heavy metals have different patterns of treatment and stimulus within a tree. Pb, Cr and Cu tend to be immobilised and held primarily in the roots, whereas Cd, Ni and Zn are more easily translocated to the aerial parts (Kabata-Pendias and Pendias, 2001). The uptake of hazardous materials, directly from wet and dry deposition by aerial parts of plants, has often been reported in the conditions of high air pollution level (Divan *et al.*, 2009).

The family Pinaceae (pine family) includes many conifers such as Cedrus, Abies, Larix, Picea, and Pinus. The Iranian Pine, *Pinus eldarica,* is famous as a Tehran pine and is a tree with wide spacing between branches and stiff long dark green needles at maturity. *Pinus eldarica* Medw. is an evergreen tree that naturally occurs in the Transcaucasian region between Europe and Asia, and grows also in Iran, Afghanistan and Pakistan (Michelozzi *et al.,* 2008). Pine needles are rich in terpenoids, polyphenols and tannins (Lee, 2003). Terpenes in conifers are significant chemomarkers of environmental impact (Supuka and Berta, 1998). Diurnal, seasonal and annual, climatic and edaphic, geographic altitudinal (Nault, 2003) variations of the concentration of terpenes in the needles of conifers have been documented.

Scientific studies about *P. eldarica* are largely restricted to botanical research. This plant consists of quercetin, pinene, myrcene, caryophyllene, camphene, and abietic acid. The main constituents of the leaf oil are germacrene D (26.6%), β-caryophyllene (17.1%), a-pinene (11.8%), elemicin (4.3%), and a-humulene (4.2%). Major components of the fruit oil are β - caryophyllene (34.0%), β-pinene (16.3%), longifolene (10.5%), a-humulene (6.4%), 3-carene (6.3%), and a-pinene (3.8%) (Afsharypour and Sanaty, 2005). Extracts of pine needles have various physiological and pharmacological actions. Experimental data relevant to the beneficial properties of *Pinus* in general relate to their anti-inflammatory (Rohdewald, 2002), antioxidant (Guri *et al.,* 2006), antineoplastic and immuno-modulatory properties (Li *et al.,* 2007).

Data concerning the effect of various anthropogenic factors on secondary compounds of the conifers is still controversial (Turtola *et al.,* 2006). Therefore, the objective of this study was, evaluation of *Pinus eldarica* needles essential oil in pollution regions

Materials and methods

Collection of plants

The needles of *Pinus eldarica* Medw. were collected in August, 2014 from different regions of Isfahan in Iran (1: Around of industrial factories. 2: In city center with air pollution. 3: Around of high voltage pylons). The plants were identified by Research Institue of Agriculture, Isfahan, Iran. The samples were separated and they were air-dried in shade at room temperature.

Gas Chromatography-Mass Spectametry (GC-MS) Analysis

The chemical composition of the needles essential oil were analyzed using GC and GC-MS. The GC/MS analysis was carried out with a 20 Agilent 5975 GC-MSD system in research laboratory of Islamic Azad University, Khorasgan Branch, Isfahan, Iran. HP-5MS column (30m × 0.25mm. 0.25mm film thickness) 20 was used with helium as carrier gas (1.2mL/min). GC oven temperature was kept 20 at 50 C2 B0C for 3 min and programmed to 280 C2 B0C at a rate of 5 C2 B0C/min, and kept 20 constant at 290 C2 B0C for 3 min, at spilitless mode. The injector temperature was at 20 280 C2 B0C. Transfer 20 line temperature 280 C2 B0C. MS were taken at 70 20 eV. Mass ranger was from m/z 35 to 450. Head space GC-MS was used in this study. This method can use plant dry matter for chemical analysis.

Results

GC-MS analysis of *Pinus eldarica* needles essential oil of collected from different regions identified 40(industrial pollution), 15(air pollution), 19(electromagnetic pollution) main compounds, respectively.

The results obtained in our study showed that major compounds of *Pinus eldarica* needles essential oil of collected from industrial region were: Octadecenoic acid (31.52%), α-pinene (14.95%), 3-carene (4.23%), β-pinene (3.151%), muskolactone (2.99%), dodecanal (2.83%) (Table 1 & Fig. 1).

The major compounds of *Pinus eldarica* needles essential oil of collected with air pollution were: α-pinene (52.57%), β-pinene (12.21%), Octadecenoic acid (7.19%), limonene (7.07%), 3-carene (4.27%), caryophyllene (4.132%) (Table 1 & Fig. 2).

The major compounds of *Pinus eldarica* needles essential oil of collected from electromagnetic region were: α-pinene (29.46%), β-pinene (12.32%), limonene (4.93%), Octadecenoic acid (4.92%), caryophyllene (2.58%) (Table 1 & Fig. 3).

Therefore, the through terpene compounds in *Pinus eldarica* needles, the α-pinene content was highest in samples collected from regions with electromagnetic pollution and air pollution, while in *Pinus eldarica* needles of collected from industrial region, Octadecenoic acid content was highest (Fig. 3, 4, 5).

Table 1. Chemical composition of *Pinus eldarica* needles of collected from different regions.

Compound	Industrial pollution (%)	Air pollution (%)	Electromagnetic pollution (%)	RT*
α-Pinene	14.95	52.57	29.46	3.596
β- Pinene	3.151	12.21	12.32	4.181
3-Carene	4.23	4.27	-	4.702

Limonene	-	7.07	4.93	4.88
Caryophyllene	-	4.13	2.58	11.56
Octadecenoic acid	31.52	7.19	4.92	21.39

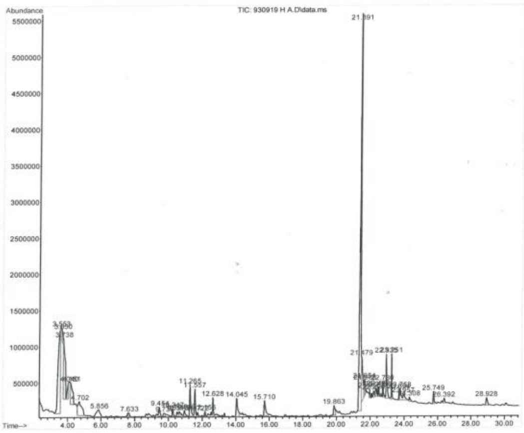

Fig. 1. Typical GC-MS chromatogram of *Pinus eldarica* needles of collected from industrial region (Data is retention time for each component).

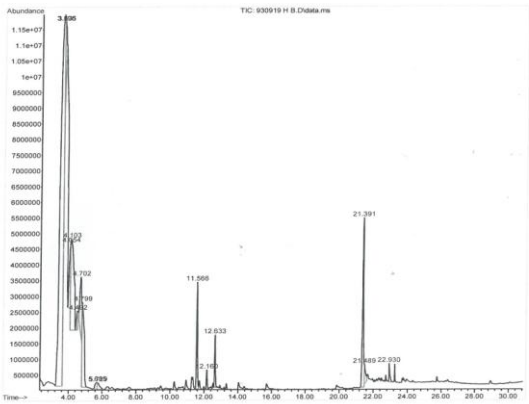

Fig. 2. Typical GC-MS chromatogram of *Pinus eldarica* needles of collected with air pollution (Data is retention time for each component).

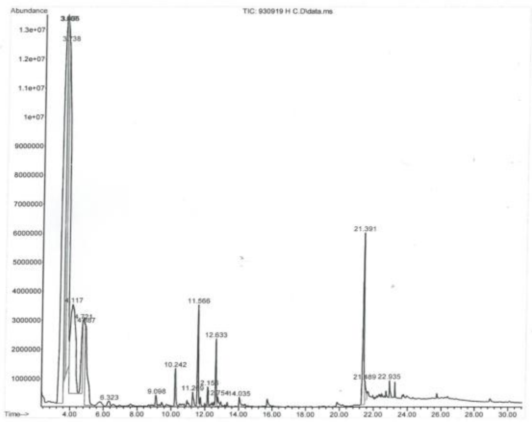

Fig. 3. Typical GC-MS chromatogram of *Pinus eldarica* needles of collected from electromagnetic region (Data is retention time for each component).

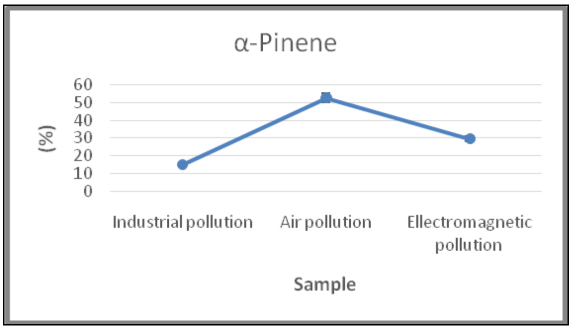

Fig. 4. Comparison of a-Pinene percent in *Pinus eldarica* needles of collected from different regions.

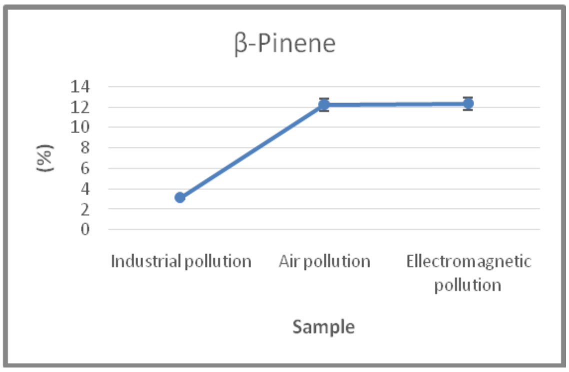

Fig. 5. Comparison of β-Pinene percent in *Pinus eldarica* needles of collected from different regions.

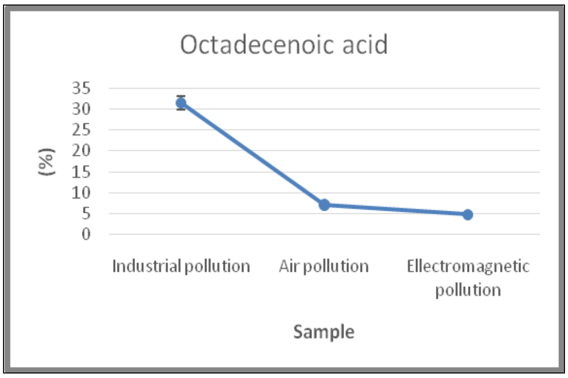

Fig. 6. Comparison of Octadecenoic acid percent in *Pinus eldarica* needles of collected from different regions.

Discussion

The results obtained in the study showed that forty, fifteen, nineteen compounds were identified in *Pinus eldarica* needles collected from industrial region, urban region and electromagnetic region, respectively. In this research was observed that the highest density of a-pinene was related to urban region *Pinus eldarica* needles but the highest densities of octadecenoic acid was obtained in industrial region *Pinus eldarica* needles.

Serbula *et al.* 2013 showed that branches and needles of pine are better biomonitors of airborne pollution with Pb than the roots of these plant species. Although Pb concentrations in the roots of pine exceeds the acute value, translocation to the aerial parts is limited. Therefore, atmospheric evidense is the most important source of this element in aerial parts. Mn concentrations in pine needles were below the suggested minimum amounts, which could be attributed to the effect of long-term air pollution with SO_2 in region and the surroundings.

Terpenes in conifers are important chemomarkers of environmental bump (Supuka and Berta, 1998). Diurnal, seasonal and annual, climatic and edaphic, geographical elatitudinal altitudinal variations of the concentration of terpenes in the needles of conifers have been attested (Nault, 2003). Data about the effect of different anthropogenic factors on secondary compounds of the conifers is still eristic (Turtola *et al.,* 2006).

So that, elevated CO_2 concentration caused an increase in the concentration of a-pinene (Salla *et al.,* 2001), thus, increased limonene emission rates in ozone-fumigated woody plants were described (Klusia *et al.,* 2002). Conifer changes in the needle a-pinene at the polluted urban site or under elevated CO_2 concentration were showed (Salla *et al.,* 2001). These results also have agreement with the research's findings.

Kurd *et al.* 2012 showed that the highest and the lowest metal concentrations were found in the heavy traffic sites and the control site, respectively. However, samples taken from highway sites includeded the high concentrations of nickel, copper and lead. Moreover, industrial areas were found to have high contents of zinc and chromium. The change in heavy metal concentrations between the studied locations is due to changes in traffic density and anthropogenic activities. This research showed significant correlations between the terpenes concentrations in pine needle samples.

Maria-Loredana *et al.* 2014 demonstrated the influence of electromagnetic irradiation at bands corresponding to wireless router and mobile devices on leaf anatomy, essential oil content and volatile emissions in *aromatic plants*. Microwave irradiation resulted in thinner cell walls, smaller chloroplasts and mitochondria, and enhanced emissions of volatile compounds, in particular, monoterpenes and green leaf volatiles. These effects were stronger for wireless router microwaves. Essential oil content was enhanced by mobile devices microwaves, but the effect of wireless router microwaves was inhibitory. These data demonstrated that human-generated microwave pollution can potentially constitute a stress to the plants. These results are similar to the results, that is environmental stresses such as electromagnetic fields effect on plant essential oils.

Conclusion

The results of this study showed that the highest density of a-pinene was obtained in urban region *Pinus eldarica* needles but the highest densities of octadecenoic acid was obtained in industrial region *Pinus eldarica* needles. The variation in terpenes concentrations between the increasing industrial and traffic activities in the city indicates the need for pollution control in the city environment. The result showed that *Pinus eldarica* Medw. needles can be used as a simple way to monitor polluted sites.

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