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EFFECTS OF AIR POLLUTION ON MORPHOLOGICAL AND ANATOMICAL CHARACTERISTICS OF *PINUS ELDARICA* WOOD

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Abstract

Air pollution, including automobile exhaust pollution, can affect anatomical and morphological characteristics of wood. In order to evaluate this subject, the *Pinus eldarica* trees of Chitgar Park in Tehran, which extends from a crowded highway in the south (polluted site) to the semi polluted midsection and to Alborz Mountain in the north (unpolluted site) were sampled with an increment borer and cores were collected. After cross dating, the tree rings of the last five years were separated from the rest of the core. Anatomical characteristics of the cross sections, including the transition between early wood and latewood, the tangential thickness of the last-formed latewood tracheids, the frequency of rays and resin ducts and the morphological characteristics of tracheids in rings formed in the same year were studied. The results indicated that the ring widths of *P. eldarica* in the three zones are not significantly different. Rays and false rings were more frequent in the polluted and semi polluted sites than in the reference area, and wall thickness was significantly different in some years. Other morphological properties did not differ significantly except for tracheid diameted. In conclusion air pollution does not alter *P. eldarica* ring width significantly but changes some anatomical and morphological properties of its wood. Therefore trees growing on polluted sites are not suitable for Dendrochronological studies.

Introduction

The relation between roadside tree growth and traffic-related air pollution has been investigated by many researchers. Some of these studies focused on effect of air pollution on leaf and bark characteristics of trees (Kurczyńska *et al*. 1997; Sarkar *et al*. 1986; Kammerbauer *et al*. 1986**;** Marmor and Randlane, 2007; Lukjanova and Mandre 2010; Li *et al*. 2010). Leaves indicate temporary pollution effects, but in contrast tree rings can permanently record environmental effects, and these effects can be detected retrospectively (Bass and Bauch 1986; Fritts 1976).

Some studies found negative correlation between ringwidth and accumulation of heavy metals like Pb, Fe, Mo, Ni etc (Lepp 1970; Baes and McLaughlin 1984, Latimer *et al*. 1996; Safdari *et al*. 2005; Medeiros *et al*. 2008; Korori *et al*. 2010). Accumulation of heavy metals in tree rings is related to soil pH (Guyette *et al*. 1991; Safdari *et al*. 2005; Augustin *et al*. 2005), and the mobility of various metals in tree boles differs (Watmough 1999) considerably.

The results of studies about the effects of other pollutant sources such as carbon dioxide, sulfur dioxide, nitrogen dioxide on the width of tree rings were contradictory (Yazaki *et al*. 2005). For example some researchers believe that when there are no limitations on environmental factors, increased carbon dioxide leads to wider coniferous tree rings (Atwell *et al*. 2003) and others found gas pollutant emissions can decrease the width of conifer tree rings (Battipaglia *et al*. 2010). Regardless, air pollution causes anatomical and morphological changes in wood texture (Donaldson *et al*. 1987; Alves 1995; Kurczyńska *et al*. 1997; Bernal-Salazar *et al*. 2004; Waris and Khan 2010).

Tehran, a heavily-populated capital, is suffering from increasing air pollution (Khorasani 1993, Safdari *et al*. 2005, Korori *et al*. 2010). Most of Iran, including Tehran, is arid with annual precipitation of 230 mm and its soils are calcareous in nature. Because of the insolubility of heavy metals such as lead, other emissions such as NOx, CO_2, CO, SO_2, fine dust particles, volatile organic compounds etc., are the main sources of air pollution (Safdari *et al*. 2005)

Softwoods are more affected by environmental conditions than hardwoods and are preferred in dendroeclogical studies. *Pinus eldarica* is one of the predominant species which have been planted in urban Tehran as a green belt for combatting air pollution and is a good candidate for a study of air pollution effects on the anatomical and morphological properties of its wood.

The aim of this research is to compare anatomical and morphological characteristics of *Pinus eldarica* wood in polluted (next to the crowded highway), semipolluted and unpolluted sites and explore its Dendrochronological potential.

Materials and Methods

Sampling sites: In order to investigate air pollution effects on the anatomical and morphological characteristics of *Pinus eldarica* wood, three sites: polluted, semi polluted and unpolluted sites were selected. Since plants are affected by climate (Fritts 1976), it was preferable to select three zones near each other.

Chitgar Park is a green belt located in the western part of Tehran City and is a vast artificial forest (950 hectares) which is connected to the polluted, crowded highway in the south and to Alborz mountain (35° 43′ 13″ N and 51° 12′ 32″ E) in the north, observable on Google Earth. The canopy of trees in the vicinity of the highway (south) is parchy while the two other sites are closed. Trees are approximately the same age and were planted 40 years ago (Safdari *et al*. 2005).

Three trees with complete canopy and straight bole and the same trunk diameter were selected from three sites. Three samples from each tree were cored with a 5 mm increment borer. The increment borer completely passed from the south side to the north side of the tree. Sampled cores were divided into two parts and the geographical directions were marked with a soft pencil.

Crossdating of core samples: To compare anatomical properties of a cross section, ring counterparts from three sites in the last five years were needed. First, the outside surface of a core sample from all trees was sanded with 100, 200, 300, and 400 grain sandpaper respectively to achieve good contrast between the border of the early and latewood. Core samples were skeleton plotted (Stokes & Smiley 1968) and crossdated, rings width were measured and a chronology was created, which is a time series of mean tree-ring width (Fritts 1976). Finally the last eight rings were separated from the rest of the core to obtain micro sections.

Softening of cores and sectioning: Softwoods with wide, thin-walled tracheids tend to collapse during sliding microtome sectioning. In order to prepare fine micro samples, all cores were kept in 4% ethylenediamine for four days, then the cores were placed in two or three changes of water at two-hour intervals to remove the ethylenediamine, and after that the wood cores were prepared for sectioning (Carlquist, 1982).

Small cores were mounted with quick-drying glue on woody core mounts and were cross sectioned by sliding microtome to prepare cross sections of 10–20 micrometers. The cross sections were dyed by safranin O and astra-blue (1:1) coloring agents then were mounted on micro slides (Parsa-pajouh and Schweingruber 1988).

After preparation of the microslides, the type of transition from early wood to latewood was recorded (IAWA committee 2004). The thickness of the tangential wall in the last two rows of tracheids (Bernal-Salazar *et al*. 2004), the frequency of rays per length and the frequency of resin canals in the rings were recorded.

Maceration of wood cores: After crossdating and measuring, the tree rings of the last five years (2003 - 2007) were cut with a scalpel. The small cores were treated with 30% hydrogen peroxide and glacial acetic acid 1:1 at 60°C for 48 h in glass-lidded test tubes. After this period the woods turned white or light yellow (Franklin, 1945). The yellow samples had been in the oven longest and their solutions were refreshed. Then the test tubes were decanted and white samples were washed in water using paper filters. After complete remove of the acid odor, the cleaned splinters were placed in a small beaker and defibered with a small magnet on an electrical magnetic plate. The length of fiber, wall thickness and lumen size of tracheids were measured with the micrometer eyepiece of a Nikon light microscope.

Data Analysis: Analysis of variance was employed to analyze the data on ringwidth and thickness of the tangential wall in the last two rows of tracheids of *P. eldarica* from the three zones for each year from 2003-2007 (SAS Institute 2000). Data on the number of rays per mm and the number of resin ducts per year were analyzed with the years 2003-2007 combined. The rings of 2003-2007 were macerated together to obtain the fibers which were measured to obtain fiber length and were used to obtain data on the morphological properties.

Results and Discussions

The mean ring width of the last five years: Mean ring width of year 2003 to year 2007 from three sites are plotted in Fig.1. Mean ring width at the unpolluted site was not significantly different from ring width at the polluted or semi polluted sites in any year from 2003-2007 (Fig. 1), although average ring width was largest at the polluted site (polluted site 3.67 mm, semi polluted site 3.59 mm and unpolluted site 3.23 mm. Our results were in agreement with Yazaki *et al* (2001), however in contrast to the finding of Atwel *et al* (2003). Different species may respond differently therefore it may be due to the difference in species in our experiments. Larger ring width in polluted sites may be due to the added CO_2 and increased temperature resulting more photosynthesis compare to the unpolluted site. All three zones had smaller mean ringwidth in the years 2004, than in the following year (2006).

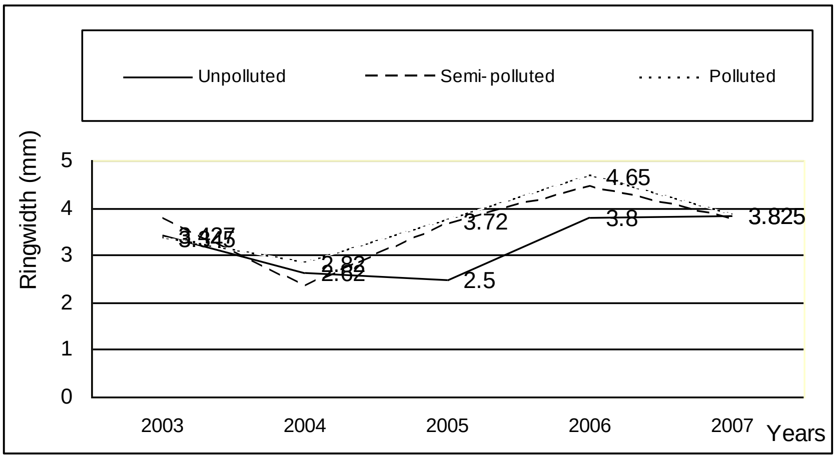

Fig.1. Mean ring width of *Pinus eldarica*

Transition from early wood to latewood: Some anatomical characteristics from year 2003 to 2007 are given in Table 1. while cells transition from early wood to latewood in different years are shown from Fig. 2a to Fig. 2e.

False growth rings occurred at the polluted and semi polluted zones during all years, except 2006 (Table 1 and Fig. 2). However in 2004 and 2005, there was one false growth ring in the unpolluted zone. Transition from early wood to latewood in all five years was gradual for the reference area and abrupt for the polluted and semi polluted zones. The present study show less number of false rings in unpolluted site.

The gradual transition of early wood to latewood in some years was affected by false rings. If a false growth ring is formed in or near the latewood, the transition from early wood to latewood seems abrupt. Since the transition in the unpolluted site is gradual, it may be conclude that pine of polluted region suffers harsh ecological conditions (IAWA Committee 2004), (Fig. 2). On the basis of these information, it may be suggested that amount of false rings increased in this species at polluted places. Kurczynska *et al* (1997) also reported the same while working on *Pinus sylvestris*. It may be anticipated that air pollution may abrupty change not only the inner condition of plant but also altered microclimatic situation around the trees, resulting abrupt change in cell characteristic (false ring), during normal annual growth period.

Table 1. Some anatomical characteristics of a cross section of *Pinus eldarica* in three zones in Chitgar Park.

Years	Number of false ring			Transition from early wood to latewood		
	Polluted	Semi-polluted	Unpolluted	Polluted	Semi-polluted	Unpolluted
2003	2	2	0	Abrupt	Abrupt	Gradual
2004	4	5	1	Abrupt	Abrupt	Abrupt
2005	5	5	1	Abrupt	Abrupt	Abrupt
2006	0	0	0	Gradual	Gradual	Gradual
2007	2	2	0	Abrupt	Abrupt	Gradual

Thickness of the tangential wall in the last two rows of tracheids: Wall thickness of annual ring of *Pinus eldarica* was shown in Fig. 3. while cell wall thickness in years 2003 to 2007 from three study areas was postulated in Fig. 4. The tangential walls in the last two rows of tracheids in the unpolluted plants were significantly thicker than the tangential wall in the polluted zone in 2003 ($F = 7.3$, $p < 0.024$) and 2004 ($F = 5.2$, $p < 0.049$) and was significantly thicker than the tangential wall in the semi polluted and polluted zones in 2005 ($F = 69.9$, $p < 0.0001$) (Figs. 3 and 4). Therefore it may be concluded that pollution may significantly reduces wall thickness at least in this species.

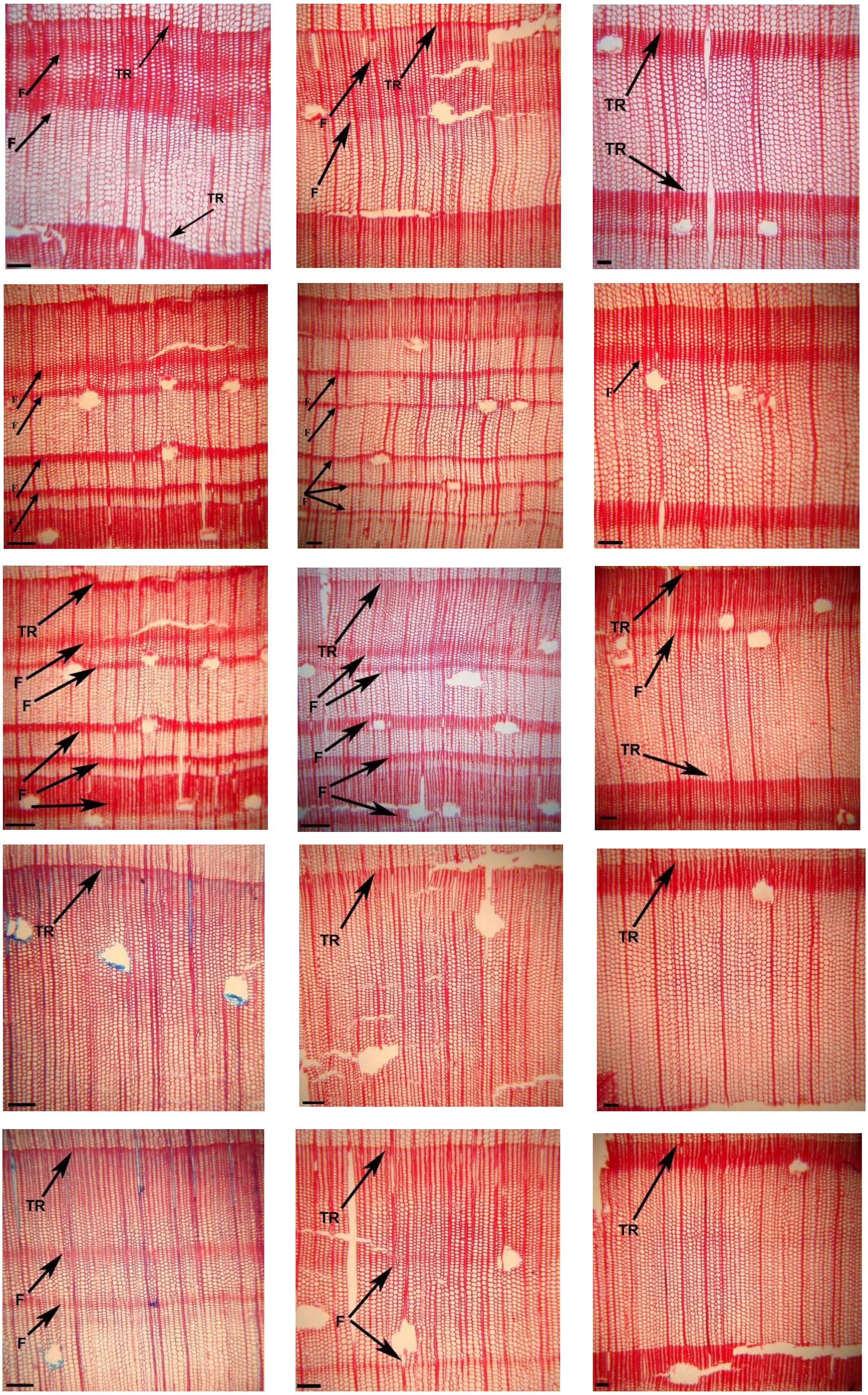

Fig.2

Fig.2b

Fig.2c

Fig.2d

Fig.2e

a-e. Cross sections of *Pinus eldarica* (from left to right: polluted, semi polluted and unpolluted). F indicates a false ring, and TR indicates a true ring. Scale bar: 200 µm. a. 2003, b. 2004, c. 2005, d. 2006, e. 2007.

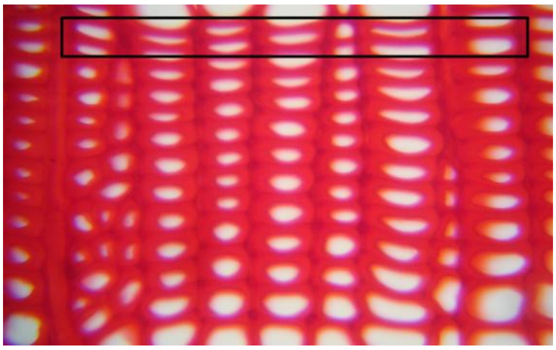

Fig.3. Light micrograph showing the last formed cell of an annual ring of *Pinus eldarica*. The rectangle indicates the cell measured

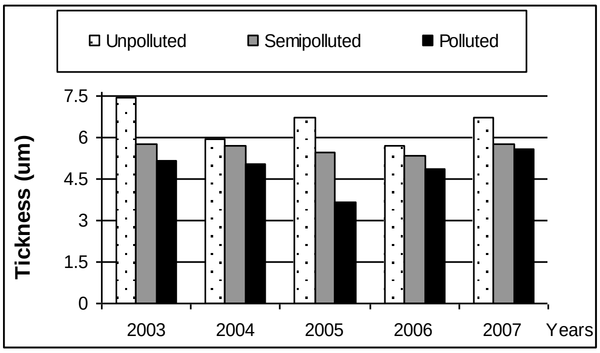

Fig.4. Tangential wall thickness of the two last formed tracheids of wood of *Pinus eldarica* in different years, in three zones of Chitgar Park)

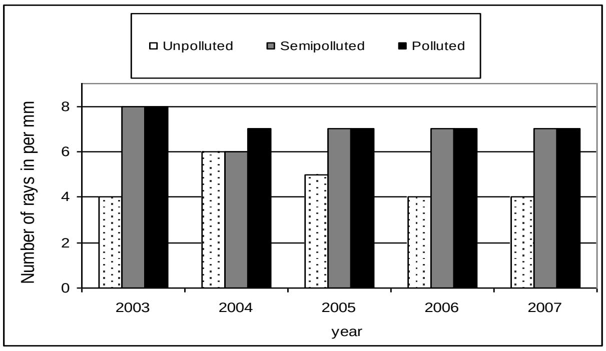

Fig.5. Number of rays per mm in wood of *Pinus eldarica* in different years in three zones of Chitgar Park

Number of rays per millimeter: Fig. 5. indicated the number of ray per millimeter in polluted and unpolluted sites. There were significantly more rays per mm in the polluted zone and in the semi polluted zone than in the unpolluted zone ($F = 20.9$, $p < 0.0001$). In every year except 2004 the number of rays in the polluted zone was equal to the number in the semi polluted zone but the number of rays was always higher in the unpolluted zone (Fig 5). Therefore it may be concluded that pollution may significantly increase the number of rays /mm in this tree species. It is indicated that anotonomical properties may change under tense situation of air pollution.

The frequency of resin ducts per year: The number of resin ducts in the unpolluted zone was considerably lower but significantly not different from other two zones in 2003, 2004 and 2006. The number of resin ducts in the polluted zone was equal to the number in the semi polluted zone in years 2003 to 2004 and 2006. It may be suggested that pollution increase the number of resins duets in *Pinus eldarica*. (Fig 6).

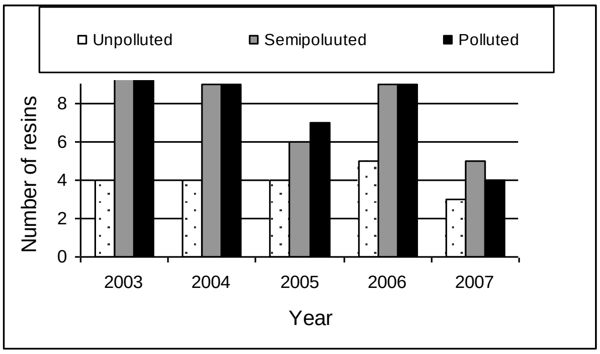

Fig.6. Number of resin ducts in wood of *Pinus eldarica* in different years, in three zones of Chitgar Park

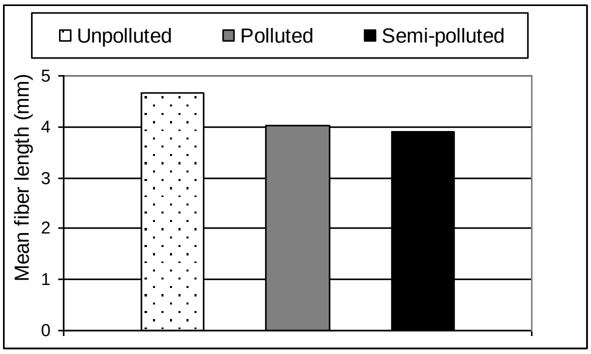

Fig.7. Mean fiber length of tracheids

Morphological properties: Fiber length, wall thickness, wall diameter and lumen diameter of *Pinus eldarica* growing in three zones are shown from Fig. 7 to Fig. 10.

Fiber length of earlywood and latewood, wall thickness of earlywood and latewood, and earlywood lumen size of tracheids were seems higher in unpolluted trees but not different significantly in the three zones (Figs. 7-9). Tracheid diameter (Fig.10) was significantly smaller in the polluted site than in the unpolluted site ($F = 5.2$, $p < 0.049$), unlike the results of Alves (1995), Bernal-Salazar *et al* (2004) and Waris and Khan (2010). Therefore it may be concluded the pollution may have adverse effect on lumen diameter in this tree species.

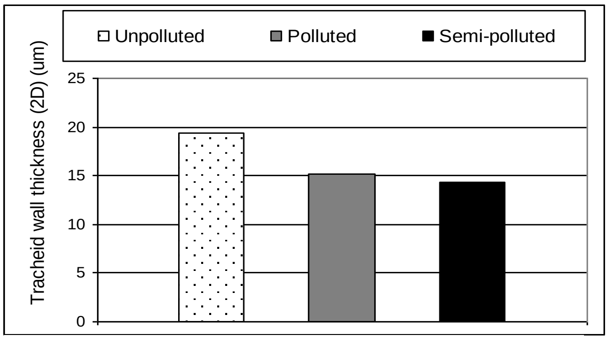

Fig.8. Mean tracheid wall thickness

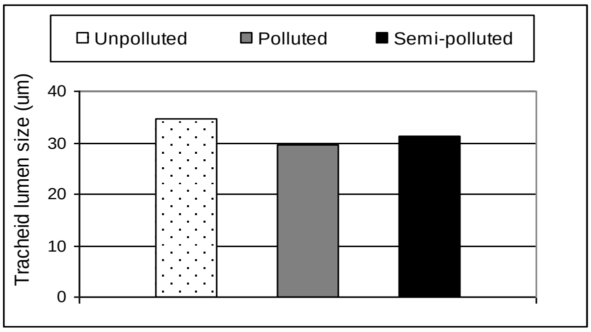

Fig.9. Mean lumen size of tracheids

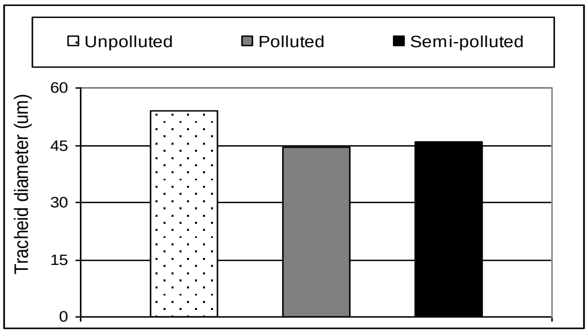

Fig.10. Tracheid diameter

Conclusions

On the basis of above studied it may be concluded that as far as *P. eldarica* is concerned.

1. Pollution did not affect mean ring width.
2. False rings were seen almost every year, but the number of them in polluted and semi polluted zones was larger than that for the unpolluted zone.
3. So air pollution, in addition to sudden change of climate, strengthens false ring formation in this species.
4. The transition between early wood and latewood in *P. eldarica* is affected by false growth rings.
5. Pollution may reduce tangential wall thickness.
6. Pollution may increase the number of rays/mm and resin ducts in this species.
7. Mean fifer length, mean wall thickness and mean lumen size of the tracheids have no significant effect of pollution while tracheid diameter significantly reduce in polluted situation.
8. The *P. eldarica* of polluted area is not ideal for dendroecological research because of false ring frequency and problems associated with cross dating especially in polluted areas.

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