Electing drought-resistant pinus pinea L. (stone pine) using dendroclimatology

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Abstract. The purpose of this study is to determine the exceptional dry years of Pinus pinea L. (stone pine) by using dendroclimatology, and Selection drought-resistant trees depending on the differentiation in the width of the annual growth rings of exceptional dry years and adopting them as seed mothers. Method. Sixteen samples were taken from four different sites (A, B, C, and D) (four samples per site). Cores were extracted from 10 trees per sample and the transverse surfaces of all cores were smoothed to expose the tree rings. Cores were then scanned and annual ring widths were measured from 2019 to 2005 using ScopeImage Plus software. By using the Moving Average as a smoothing technique, the exceptional years of the time series for the width of the annual growth rings for each sample were determined. More specifically, results show that 2008 and 2014 are negatively exceptional years across all the sites. Furthermore, the correlation coefficients between the ring-widths, the annual precipitation, and temperature for each site show that the annual precipitation is the determinant of the occurrence of exceptional years and that 2008 and 2014 are the dry years in our chosen sites. When comparing the ring widths of the dry years, it was noticed that trees (Aa4, Ab6, Ac9, Ad2) in the site A, (Ba10, Bb1, Bc3, Bd1) in the site B, (Ca9, Cb5, Cc8, Cd1) in the site C and (Da1, Db3, Dc7, Dd3) in the site D are superior and have wide rings. Scientific novels. According to this study, these trees can be selected as drought-resistant and adopted as seeds mothers which can be used in the propagation of Pinus pinea.

1. Introduction
A drought is considered one of the most important challenges facing plant breeders and harming the local economy. It can have a substantial impact on the ecosystem and agriculture of the affected region [1, p. 1]. Drought affects the growth of trees, therefore the drought-resistant plant species will be viable [2, p. 246].

A higher frequency of increasingly severe droughts highlights the need for short-term measures to adapt existing forests to climate change [3, p. 1].

Selection is the first genetic improvement processes, this process means that a very small percentage of the population for one or more of the desired traits will be selected. The selection of an adapted population and seed trees inside them represent a chance to increase forest stability and productivity over the next generations of breeding [4, p. 2].

Conifers are one of the main types that form the natural forests. On a global scale, the family Pinaceae far outweighs all other conifers in economic importance; among the genera in this family Pinus ranks first [5, p. 12].
Umbrella pine (Pinus pinea L.) (Pinaceae), is widely distributed throughout the Mediterranean basin, where it is native, covering more than 700,000 ha as the dominant species [6, p.1], with a recent increasing expansion owing forest restoration or farmland afforestation [7, p.426]. Stone pine (Pinus pinea L.) is a Mediterranean tree. It is well suited to high temperatures and drought [8, p. 563].

Pinus pinea L. is one of the most characteristic tree species of the landscape because of its singular umbrella shape and the ancient use of its large, nutlike edible seeds for human consumption [9, p. 2].

Stone pine is frequently preferred in afforestation practices due to its ecological, economic, and aesthetic characteristics, and it is among the top species that provide an important contribution to the national economy as a non-wood product [10, p.416].

Dendrochronology can be defined as the study and reconstruction of past changes that impacted tree growth [11, p. 1].

Dendrochronology is one of the methods applied in reconstructing past environmental events. It is the science of defining past climatic and hydrologic variability from tree-ring data [12, p. 1].

Dendroclimatology is one of the subdisciplines of Dendrochronology, it uses the information in dated rings to study problems of present and past climates [13, p. 139].

Dendroclimatology is concerned with constructing records of past climates and climatic events by the analysis of tree growth characteristics and especially annual rings [14, p. 175].

In certain areas, soil moisture declines when precipitation decreases and when the depth of groundwater increases [15, p. 55],[16, p. 377]. Therefore, trees will produce wider annual rings in wet years compared to dry years. Therefore, tree rings and vessel features are valuable proxies in the evaluation of climatic changes.

Tree annual growth rings are considered an indirect source of information about changes in the environment. The exceptional changes of environmental conditions cause the occurring of exceptional growth rings. Either the growth rings are wider (wider when conditions favor growth) or the growth rings are narrower (when times are difficult) by comparing with the annual adjacent growth rings on the cross-section of the tree trunk [17, p. 245].

For example, when an exceptional growth event is detected (whether it is a wider growth ring or narrower growth ring in a specific year compared to previous and next years) in the same year for all trees within the same forest, the year could be called an exceptional year [18, p. 1].

In the study [19, p. 22] "Influence of Precipitation and Temperature on Tree-Ring Width of Brutia Pine (Pinus brutia Ten.)", the researcher deduced that by using Dendroclimatology tree-ring width was more influenced by precipitation than temperature, especially in growing season and prior to it.

Ring-width chronologies of P. l察teri at three sites showed significantly positive correlations with precipitation, relative humidity, and self-calibrated Palmer Drought Severity Index (scPDSI) during the dry season (previous November to current April) and early rainy season (May–June). Conversely, significantly negative correlations were found between ring-width site chronologies and air temperatures (mean, maximum, and minimum) from April to August [20, p. 1].

It was used a tree-ring database to study how drought influences forest growth resilience [21, p. 1].

Based on data of tree growth rings, it was described the black pine specific climate-tree-growth relationship and the vitality response of Black pine to droughts [22, p. 1].

The study [23, p. 32] was intended to estimate growth changes and to develop an estimating formula for ring-growth based on climate factors for Quercus mongolica by using Dendroclimatology.

Increasing drought is one of the most critical challenges facing species and ecosystems worldwide, and improved theory and practices are needed for quantification of species tolerances [24, p. 1].

Forest management practices can partially ameliorate drought impacts through the selection of drought-tolerant species and genotypes [25, p. 2329].

Climate projections forecast drier and hotter conditions and an increasing trend in extreme climatic events such as drought. Scientific evidences reported that extreme dry spells affected the stem growth of different Mediterranean low-elevational pine forests inducing a decrease in tree resilience, defined as the capacity to resist environmental stress and to recover pre-disturbance functioning [2, p. 246].
In this light, the purpose of the research is to determine the exceptional dry years of Pinus pinea L. (stone pine) by using dendroclimatology, and Selection drought-resistant trees depending on the differentiation in the width of the annual growth rings of exceptional dry years and adopting them as seed mothers (source of seeds).

2. Methods
There are 4 different sites were selected in forest Pinus pine (A, B, C, D), four samples were selected within each site, the area of one or repeated sample is $400m^2$. These selected samples are representative of the whole site.

Ten trees were selected to extract the cores (5 trees were phenotypic superior trees, other 5 trees each of them represents several similar trees which they had the same phenotypic traits.) i.e. 10 trees per sample, so the number of trees (which were extracted the cores) were 40 trees for each site (A, B, C, D), the total numbers of trees for this study were 160 (at the four sites).

Samples were taken by increment borer with sample long (20 cm), three cores were extracted at breast height (130 cm) across three directions (N, SE, and SW).

After extracting the samples, they were fixed on cardboard, which was prepared to hold the samples as shown in figure (1).

After samples were dried, we put them on a special wooden stand with ducts as shown in figure (2).

After samples were completely dried, samples were smoothed by coarse sandpaper, then using fine sandpaper they were smoothed many times until edges of growth rings were clear, they can be seen with naked eyes as shown in figure (3).

![Figure 1. Fixing the cores on temporary stands (cardboard).](image1)

![Figure 2. Fixing the cores on permanent stands (wooden stand).](image2)
Dendrochronology (or tree-ring dating) is the scientific method of dating tree rings (also called growth rings) to the exact year they were formed. As well as dating them this can give data for dendroclimatography, the study of climate and atmospheric conditions during different periods in history from wood. Dendrochronology is useful for determining the precise age of samples by comparing the growth rings of samples, firstly it should be compared the samples from the same tree, then comparing these samples with the other samples from the other trees [11, p. 1].

Measuring the width of the annual growth rings: firstly samples were scanned, the images were handled with the ScopeImage Plus program later they were zoomed 300% to measure the width of each growth rings by using the ruler of the program as shown in figure (4), after getting the width of the growth rings in pixels, it should be converted into mm. The measurement process was carried out from the 2019 ring to the 2005 growth ring, where 15 growth rings were measured the width for each sample at all sites.

Determining the exceptional years: it could be determined the exceptional years of the time series for the width of the annual growth rings for each sample by using the moving average in the Excel program [26, p. 87]. The exceptional growth response which is observed within a time series for one tree within the studied sample is known as "Event year", this year can also be called the exceptional year (Pointer year) where the annual event year is repeated in all samples of trees.

* the first stage: the annual events were calculated by using the moving average, where the length series of subsets (of the full data sets) was 3 years for each series of the width of annual growth rings using the following relationship[27, p. 5]:

![Figure 3. The cores after using fine sandpaper.](image)

![Figure 4. Measuring tree ring-width with ScopeImage Plus.](image)
\[ z_i = \frac{x_i - \text{mean}[\text{Window}]}{\text{stddev}[\text{Window}]} \]  

(1)

\( z_i \): value the annual event in year \( i \).
\( x_i \): the growth ring width in year \( i \).
[Window] mean: the average width of the growth rings during the selected window (subset with length 3 years).
[Window] stdev: The standard deviation of the growth rings width within the selected window (subset with length 3 years).

Negative and positive annual events were determined from \( z_i \) threshold, so that they were:
- \(-1 \leq z_i\): negative annual event.
- \(1 \leq z_i\): positive annual event.

It should be pointed out that this study just focused on the negative events.

* the second stage: the negative exceptional year was determined as followed: the year is considered a strong negative exceptional year (sNPY) when more than 50% of negative annual accidents are registered within the time series for the width of growth rings in each sample, while the year is considered a medium negative exceptional year (mNPY) When 25-50% of annual accidents are registered within the time series of growth rings width in each sample.

To explain the occurrence of exceptional years, it was used climate data (monthly total precipitation, monthly mean maximum temperatures, monthly mean minimum temperatures, and monthly medium temperatures of the weather stations close to the study sites.

**Election drought-resistant trees:** After measuring the width of the annual growth rings for all the cores which already had been extracted (the three cores for each tree) and measuring the width of the annual growth rings for all the trees (which were extracted the cores), the average of the three cores was calculated so there is a value that represents the average width of the ring corresponding to the year (15 values represent the average width of the ring corresponding to 15 years). After determining the exceptional years, it was separately compared the values the width of the ring corresponding to these exceptional years within each sample, so it was deduced the ring with the maximum value of the width (the widest) which indicates the tree resistance to the exceptional condition represented by drought (that is done for each sample).

Finally, for each sample, it was elected the tree which had the widest ring in the exceptional year and it was considered a drought-resistant tree.

3. Results

1. Exceptional years:

Site (A): It was observed that there were four strong negative exceptional years 2008, 2014, 2018, 2011.

Site (B): four exceptional years were observed at the site (B), two years were strong negative ones 2008, 2014, and two medium negative ones 2011, 2018.

Site (C): it was observed that there were five exceptional years, two were strong negative 2008, 2014, and three medium negative years 2011, 2018, 2005.

Site (D): While there were three exceptional years in the site (D), two were strong negative 2008, 2014, and a medium negative one 2009.

When it was compared the four studied sites, it was found that the years 2008, 2014 are strong negative exceptional years for all sites. Therefore, it can be considered these years which the values of the growth ring width (corresponding to these years) will be compared for each sample, and all the sites.

2. Study the correlation between annual growth rings width and climatic factors (precipitation - temperature): to explain climatically the occurrence of exceptional years, the correlation coefficient was calculated between the series of growth rings for each site and the time series of total annual of precipitation and annual mean temperature (low - maximum - medium) during
the study period (2005-2019). It was used SPSS to calculate the Pearson correlation coefficient values as shown in table (1).

**Table 1.** The values of the correlation coefficient between the series of growth rings for each site and the time series of annual total of precipitation and annual mean temperature (low - maximum - medium)

| Site | Precipitation | Maximum temperature | Low temperature | Medium temperature |
|------|---------------|---------------------|-----------------|-------------------|
| A    | 0.78**        | 0.022               | -0.323          | -0.233            |
| B    | 0.691**       | 0.138               | -0.276          | -0.158            |
| C    | 0.874**       | 0.206               | -0.132          | -0.045            |
| D    | 0.91**        | 0.128               | -0.294          | -0.225            |

*Note: ** indicate the significance of the Pearson correlation coefficient.*

By looking at the values of the correlation coefficient, it was noted that there is a strong positive correlation between the annual precipitation and the width of the annual growth rings, while the correlation was weak negative and insignificant between the average of the annual temperature (low and medium) and the width of the annual growth rings, and the correlation was positive, weak, and insignificant between the average of the annual temperature (maximum) and the width of the annual growth rings.

According to these results, it can be said that the annual precipitation is the determining factor of the occurrence of the exceptional years, by reference to the annual precipitation data of two weather stations for the exceptional years 2008, 2014 it was found that the annual precipitation averages for these years were the lowest by comparing the other years of study, as shown in table (2).

Therefore, it can be said that the years 2008, 2014 are the exceptional years of drought for the study sites. So, it can be adopted to compare the width of growth rings for these two years among the individuals of the same sample to select the best rings (the widest), which represents the drought-resistant tree.

**Table 2.** The annual precipitation of the exceptional years and the average precipitation of the study years.

| The weather station | The precipitation (mm) |
|---------------------|------------------------|
|                     | 2008       | 2014    | Average |
| Station1            | 259        | 266.5   | 448.5   |
| Station2            | 176        | 208.5   | 279.6   |

3. Election drought-resistant trees based on the differentiation of width of annual growth rings for exceptional dry years:

3-1 Site (A): It can be noticed the superiority of trees (Aa4, Ab6, Ac9, Ad2) over the other trees in the site(A) as shown in figure (5), where was found the difference in the width of their growth rings within exceptional dry years, although there were the similar environmental conditions at the sample, therefore it can be considered these trees are genetically superior in drought resistance over the trees within the same sample, consequently it must be elected and adopted as drought-resistant seed mothers.
3-2 Site (B): The results of the comparison of the annual growth rings width of the individuals at the site (B) in each of the four selected samples showed that the trees (B10, B1, B3, B11) were superior over the other trees in both the exceptional dry years as shown in the figure (6).

Although the drought was exceptional, the difference of growth rings width is a strong indicator that these trees had genes of drought-resistance, therefore they can be adopted as drought-resistant elected seed mothers at the site (B).

**Figure 5.** Average width of annual growth rings of Site (A) in 2008, 2014.

**Figure 6.** Average width of annual growth rings of Site (B) in 2008, 2014.
3-3 Site (C): It can be noticed the superiority of trees (Ca9, Cb5, Cc8, Cd10) over the other trees in the site (C) as shown in figure (7), where was found the difference in the width of their growth rings within exceptional dry years, although there were the similar environmental conditions at the sample, therefore it can be considered these trees are genetically superior in drought resistance over the trees within the same sample, consequently it must be elected and adopted as drought-resistant seed mothers.

![Figure 7. Average width of annual growth rings of Site (C) in 2008, 2014.](image)

3-4 Site (D): The results of the comparison of the annual growth rings width of the individuals at the site (D) in each of the four selected samples showed that the trees (Da1, Db3, Dc7, Dd3) were superior over the other trees in both the exceptional dry years as shown in the figure (8). Although the drought was exceptional, the difference of growth rings width is a strong indicator that these trees had genes of drought-resistance, therefore they can be adopted as drought-resistant elected seed mothers at the site (D).
Figure 8. Average width of annual growth rings of Site (D) in 2008, 2014.

4. Conclusion
1- The exceptional years of the time series (2005-2019) for the width of the annual growth rings of Pinus pinea L. (stone pine) at the sites (A, B, C, D) were determined by using the moving average, it was found that the years 2008, 2014 were the exceptional years, both were strong negative exceptional years for all the sites.
2- It was calculated the correlation coefficients for the growth rings for each site and the series of total annual precipitation and annual mean temperature (low-maximum-medium), these values of the correlation coefficients were showed that the variable annual precipitation was the determining factor of occurrence of the exceptional years, there were two exceptional dry years for all the studied sites they were 2008, 2014.
3- After comparing the width of the annual growth rings of the exceptional dry years 2008, 2014 in the studied sites, the results showed that there were four trees per site which were superior over the others. Hence it can be considered these trees are genetically superior in drought resistance over the trees within the same sample, consequently it must be elected and adopted as drought-resistant seed mothers of Pinus pinea L. (stone pine) and using it to propagate the species.

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