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Nutraceutical and nutritional characteristics of capulin segregants (*Prunus serotina*) fresh and processed

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Abstract

The capulin fruit (*Prunus serotonin;* Family *Rosacea)* has been valued since pre-hispanic times for its medicinal properties, used in the treatment of some diseases. Although Mexico is part of the center of origin of the capulin, production and consumption of this fruit have decreased in recent years, becoming an underutilized fruit. There is little research on its nutritional and nutraceutical properties. The aim of this investigation was to evaluate the physicochemical properties, nutritional and nutraceuticals components of fresh and processed capulin fruits, from four segregants. Polar and equatorial diameter, peel color was determined by evaluating *L* (brightness), the angle of tone (*hue)* and color purity or *chromaticity* index (*chroma*), pH, and TSS; as well as the content of carbohydrates, ash, humidity, crude fiber, protein, and lipid content were quantified according to the AOAC guidelines. Mineral content was quantified by atomic emission spectrophotometry, phenolic compounds by the Folin-Ciocalteu method, anthocyanins by the pH differential method, and antioxidant activity by the ABTS method. The fruits showed high protein and fiber contents. Significant differences in nutraceutical content were found among the four types of segregants. The thermal process did not decrease the nutraceutical quality (except anthocyanins) of the four types of segregants, this only affected the nutritional attributes. Therefore, the segregants with the highest nutraceutical value were Puebla 5-28F and Puebla 5-3F, due to their high contents of phenolic compounds and anthocyanins. In conclusion, capulin fruits contain a wide variety of antioxidant and nutritional compounds, and their consumption could generate benefits for human health.

Keywords: antioxidants, minerals, proximal, genetic segregation.

Introduction

Recently, in Mexico there has been a growing interest in the knowledge and management of underutilized fruit, also known as minor, secondary or alternative fruits, as it is the case of the capulin *(Prunus serotina)*. Capulin belongs to the family Rosaceae and to the gender *Prunus,* where more than 200 species of commercially important species are found, such as cherry, peach, plum, among others, known as stone fruits (Potter, 2011). In 1951, McVaugh described five subspecies of *Prunus serotina,* the subspecies *capuli, serotina* and *virens,* coexist in several states of Mexico (Guzmán et al., 2020).

Prunus serotina is a deciduous tree native to America which grows in diverse regions in the wild or cultivated conditions, in cool semi-cold, humid and temperate climates. Its distribution includes southeastern Canada, northeastern United States, Ecuador, Colombia, Guatemala and the Sierra Madre Oriental, Occidental and the Neovolcanic axis of Mexico, (López-Hernández et al., 2024; Pathania et al., 2022). Currently, this species is naturalized in several countries around the world, including in various regions of Europe (Germany, Denmark, France, England, Lithuania, the Netherlands, Poland, Romania, Switzerland, among others) (Petitpierre et al., 2009). In the United States,

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capulin is known as wild or black cherry, in Europe as Mexican cherry (Petitpierre et al., 2009). In 2023, Mexico reported formal cultivation of capulin only in Veracruz, Mexico City, Puebla, Estado de Mexico and Jalisco, with a harvested area of 37 ha with a production of 114.28 t of this fruit (SIAP, 2024).

Capulin fruits are globose fleshy drupes, reddish to black in color depending on their state of ripeness, climacteric in nature and sweet and sour in taste; they also commonly contain cyanogenic glycosides (prunazine and amygdalin), excessive consumption, without thermal processing, can have adverse health effects (Telichowska et al., 2020). According to Swain et al. (1992), the presence of cyanogenic glycosides in some members of the genus *Prunus* is considered a defense mechanism of the plant against herbivores and pathogens through the release of hydrogen cyanide (HCN) and benzaldehyde. In the particular case of *P. serotina,* it is reported to accumulate high levels of cyanogenic glycosides in the ripe fruit; However, it lacks the enzymes amygdalin hydrolase (AH), prunasin hydrolase (PH) and mandelonitrile lyase (MDL) that by hydrolysis would release HCN, so these only contribute to the bitter taste that is compensated by the accumulation of sugars at maturity of consumption, in contrast, the seeds during the roasting process are destroyed by the temperature of the cyanogenic glycosides (Telichowska et al., 2020) .

In general, the fruits of *P. serotina* are marketed fresh, dried, or in jams, liqueurs or syrups; the seeds are eaten roasted with salt as a snack (Ordaz-Galindo et al., 1999). Since pre-Hispanic times, its fruits have been traditionally used for the treatment of some diseases (respiratory, cardiac, stomach and hypertension) (García-Aguilar et al., 2015; Luna-Vázquez et al., 2013). In addition, the capulin fruit has attracted attention as a potential source of nutrients and antioxidants. Ordaz-Galindo et al. (1999) reported the presence of anthocyanins (cyanidin-3-glucoside and cyanidin-3-rutoside) in the peel of *P. serotina* subsp. *capuli*. Moreover, Ibarra-Alvarado et al. (2009) point out the presence of antihypertensive compounds such as some phenolic ones (acid chlorogenic) in the fruit, metabolites that could justify its medicinal properties. Hernández Rodríguez et al. (2019) report that the content of flavonoids and phenolic compounds decreases in the last stages of fruit ripening, with a considerable increase of total anthocyanins of up to 1.4 mg cyanidina-3-glucósido $\cdot g^{-1}$ dry weight. The high content of phenolic compounds (phenolic acids), flavonoids (anthocyanins, proanthocyanidins, catechins), essential oils and tannins (Jiménez et al.; 2011.; Luna-Vázquez et al., 2013) explain its use as a natural therapy for the treatment of neurodegenerative diseases, such as some types of cancer, immune system problems and cardiovascular diseases (Potì et al., 2019; Telichowska et al., 2020)). In addition, capulin seed is a significant source of minerals, unsaturated fatty acids (oleic, linoleic and α-eleostearic acid) and high-quality, highly bioavailable proteins (García-Aguilar et al., 2015).

On the other hand, genetic studies of *P. serotina* have reported the phenomenon of allopolyploidy (union of genomes from different species), so intraspecific hybrids of capulin in Mexico with characteristics of different subspecies are available (Fresnedo-Ramírez et al., 2011; Pairon & Jacquemart, 2005). In addition, the morphological variability of the capulin in central-western Mexico is a product of human selection, directed at anthropocentric characters of interest, so that fruits of several sizes, degree of sweetness, with epicarp from reddish to almost black colorations, are found in wild populations, in *in situ* managed and cultivated (Fresnedo-Ramírez et al., 2011; Guzmán et al., 2020).

In this context, the morphological variability of the capulin is useful for the genetic improvement of this species, aimed at obtaining selections with better fruit and seed quality, or nutritional and nutraceutical value. In this sense, the Colegio de Postgraduados has a capulin collection with several segregant lots derived from outstanding individuals from the state of Puebla, mainly for their physicochemical characteristics. The term segregant used in this research denominates to individuals that were born by growing seeds from the same tree (sexual reproduction). Therefore, the objective of this study was to evaluate the physicochemical characteristics, content of nutraceutical compounds, antioxidant capacity and nutritional composition in the fruits of four fresh and processed capulin segregants.

Materials and Methods

Plant Material

Fruits were collected at commercial maturity stage from four segregants (individuals born from different seeds of the same tree) of capulin (*Prunus serotina)* grown in the Fruit Orchard of the Colegio de Postgraduados, Campus Montecillos, municipality of Texcoco, Estado de México, México (19°27’ N, 98°54’ O, 2 245 msnm): Puebla 5-1 (P5-1F), Puebla 5-3 (P5-3F), Puebla 5-18 (P5-18F) y Puebla 5-28 (P5-28F). The four segregants were selectected because they are offshoots of the same tree, under the same edaphoclimatic conditions.

Experimental Design

The physicochemical characterization of the fruits of four capulin segregants was carried out under a completely randomized experimental design with 25 replications, the experimental unit consisted of one fresh fruit with seed. The mineral content in the pulp of fresh fruits of four capulin segregants was carried out under a completely randomized design with three replications, considering 100 g of fruits with peel and without seeds of each fresh segregant as the experimental unit. The effect of thermal treatment on the proximal and nutraceutical characteristics of capulin fruits was evaluated using an asymmetric factorial experimental design with completely randomized allocation for the study of the factors: capulin segregant (four segregants) and degree of processing (fresh and processed). A total of eight treatments were evaluated with three replications. The experimental unit was 100 g of capulin fruits with peel and seedless of each segregant, fresh or processed (Table 1).

Statistical analysis

The data were submitted to an analysis of variance (ANOVA) and Tukey's mean comparison test ($P < 0.05$), using the Statistical Analysis System program (SAS Institute Inc., 2002). The results of the evaluated variables were expressed as the mean standard deviation.

Sample preparation

For processing, the fruit in water was kept at 40 °C by 5 min in an electric grill (Corning, model PC-620D, USA). The processed and fresh fruits were frozen with liquid nitrogen and stored to -18 °C, until analyzed.

Physical-chemical characterization

Polar and equatorial diameters of 25 fresh fruits with peel and seed of each segregant were measured using an electronic caliper (Truper, model CALDI-6MP, Jilotepec, Mexico). Fruit weight was determined on an analytic balance (Adventurer Pro AV64C, Ohaus Corporation, New Jersey, USA). Similarly, these variables were measured in the pulp-free capulin seed.

Peel color of fruit from each segregant was determined by evaluating *L* (brightness), hue angle tone (*hue*) and color purity or chromaticity (*chroma*) with a digital colorimeter (Chroma Meter CR-400, model B8210363, Konica Minolta Sensing, Inc., Tokyo, Japan) as described by McGuire (1992).

Total Soluble Solids (TSS) were determined using a refractometer (Hand-Held Refractometer, N-1E, ATAGO, Tokyo, Japan) and pH using a potentiometer (HI2211 pH/ORP Meter, Hanna Instruments, Woonsocket, RI, USA) as established by the AOAC (2005).

Quantification of minerals

Peeled and seedless capulin fruit from each segregant were dehydrated in a forced convection air oven (Binder®, model KB115 Tuttlingen, Germany) to 60 °C by 48 h. The dried and milled samples were subjected to di acid wet digestion diácida (H_2SO_4:$HClO_4$, 4:1 v/v and H_2O_2) in a Digestor™ (Tecator Kjeltec FOSS, model DT 220, Hoeganaes, Sweden). The determination of B, Ca, Cu, Fe, K, Mg, Mn, Na, P, and Zn was performed according to the methodology described by Alcántar-González & Sandoval-Villa (1999) in an Induction Coupled Plasma – Atomic Emission Plasma Spectrophotometer (ICP-AES, Instrument Varian Liberty series II, Sydney, Australia).

Proximal analysis

Carbohydrate, ash, humidity, crude fiber, protein and lipid contents were determined according to AOAC (2005). The results were expressed as percentage of fresh weight.

Quantification of nutraceuticals

Preparation of methanolic extract. To 1g of fruit pulp with peel from each segregant (fresh and processed) was added 10 mL of aqueous MeOH to 80 % (v/v), the mixture was homogenized by shaking in a vortex (Barnstead International, model M16715, Iowa, USA). Subsequently, it was sonicated (Cole Parmer 8892, Illinois, USA) for 15 min at environment temperature and allowed to settle for 24 h. Finally, it was centrifuged (Cole-Parmer Instrument Company, model 8892, Vernon Hills, IL, USA) at 1 409 g (10 min) for nutraceutical quantification (Román-Cortés et al., 2018).

Determination of phenolic compounds. 0.5 mL of the methanolic extract was taken and 0.5 mL of Folin-Ciocalteu reagent (0.2N) and 4 mL of 0.7 M of Na_2CO_3 were added,

Table 1. Physical attribute of fruits and seeds of four segregants of capulin (*Prunus serotina*).

Variable	Segregant			
	P5-1F	P5-3F	P5-18F	P5-28F
	Fresh fruit with seed			
Weight (g)	3.04 ± 0.53 b	3.39 ± 0.32 a	2.18 ± 0.28 d	2.73 ± 0.26 c
Equatorial diameter (mm)	17.58 ± 1.27 b	18.83 ± 0.78 a	16.51 ± 0.86 c	16.74 ± 0.57 c
Polar diameter (mm)	15.69 ± 0.78 b	16.29 ± 0.45 a	14.76 ± 0.60 c	15.07 ± 0.52 c
	Seed			
Weight (g)	0.38 ± 0.03 b	0.51 ± 0.06 a	0.32 ± 0.02 c	0.38 ± 0.02 b
Equatorial diameter (mm)	9.57 ± 0.26 b	10.72 ± 0.29 a	9.09 ± 0.28 c	9.48 ± 0.26 b
Polar diameter (mm)	10.84 ± 0.40 c	12.38 ± 0.45 a	10.08 ± 0.32 d	11.77 ± 0.38 b

Values represent the average of 25 replicates ± standard deviation. Means with the same letter, in the same row, are statistically equal (Tukey, 0.05).

the mixture was incubated at environment temperature in darkness by 2 h. Readings were taken on a UV/Vis spectrophotometer (Thermoscientific, Genesys 10s, Florida, USA) at 765 nm. Concentration was calculated from a standard curve ($y = 0.0068x – 0.0003$; $R^2 = 0.995$) based gallic acid (Waterman & Mole, 1994). Total phenolic content was expressed as mg gallic acid equivalents per 100 g of fresh weight (mg EAG·100 g^{-1} p.f.).

Quantification of flavonoids. It was carried out following the method reported by Chang et al. (2002). To 0.5 mL of methanolic extract was added 1.5 mL of methanol (95 %), 0.1 mL de $AlCl_3$ (10 % p/v), 0.1 mL 1 M de CH_3COOK y 2.8 mL of distilled water. The mixture was homogenized and incubuated for 30 min to environment temperature in darkness. The absorbance was read at 415 nm in a UV/Vis spectrophotometer. The standard curve ($y = 0.007x – 0.0051$; $R^2 = 0.999$) was constructed based on quercetin. Results were expressed as mg quercetin equivalents in 100 g fresh weight (mg EQ·100 g^{-1} p.f).

Quantification of anthocyanins. It was carried out by means of the differential pH method described by Giusti & Wrolstad (2001). Two 0.2 mL samples methanolic extract were taken; to the first one 1.8 mL of a buffer solution pH = 1.0 (KCl) was added to the second one a buffer solution pH = 4.5 ($CH_3COOH/CH_3COONa \cdot 3H_2O$) was added. Both samples were measured for absorbance at 510 and 700 nm. The total absorbance (A_T) was calculated from the formula: $At = [(A_{510} – A_{700})_{pH = 1.0}] – [(A_{510} – A_{700})_{pH = 4.5}]$. The anthocyanin concentration was calculated by the equation: Anthocyanins($mg \cdot L^{-1}$) = (A_T * PM * FD * 1000) / (ε * 1); where: A_T=total absorbance, P.M = molecular weight (449.2 g·mol^{-1}) of cyanidin-3-glucoside, FD = dilution factor (10), ε = molar absorptivity of the standard (26 900). Concentration was expressed as mg cyanidin-3-glucoside per 100 g fresh weight of capulin.

Quantification of vitamin C (ascorbic acid). It was determined in pulp with peel of fresh and processed fruits of the four segregants, following the methodology described by Dürüst et al. (1997). For the preparation of the extract, 1 g of plant material was placed in 10 mL of 0.4 % (p/v) oxalic acid. The mixture was sonicated for 15 min at environment temperature, then filtered. One mL of the extract was mixed with 1 mL of acetate buffer pH = 3 (3 g anhydrous sodium acetate in 7 mL water and 10 mL glacial acetic acid) and 8 mL dichloroindophenol (from a 12 mg·L^{-1}aqueous solution), after 15 s, absorbance was measured at 520 nm in a spectrophotometer. Results were expressed as mg ascorbic acid per 100 g of fresh weight (mg EAA·100^{-1} p.f.), using an ascorbic acid standard curve ($y = 0.004x + 0.0011$; $R^2 = 0.997$).

Evaluation of antiocidant capacity. To 10 mL solution of the ABTS radical $^{\bullet+}$(acid 2,2'-azinobis (3-ethylbenzzothiazolin)-6-sulfonic acid), 6.61 mg of $K_2S_2O_4$, was added, the mixture was allowed to stand at environment temperature in darkness for 16 h (Re et al., 1999). 1 mL of the ABTS radical was taken and absolute ethanol was added until an absorbance of 0.7 ± 0.01 a was obtained at a wavelength of 734 nm. To 1 mL of the ABTS radical, 10 µL of the extract to be analyzed was added and the mixture was incubated at 30 °C in darkness for 7 min. Finally, absorbance reading at 734 nm was taken. A standard curve ($y = -0.2895 x + 0.7583$; R2 = 0.9956) was prepared based on trolox. The results were expressed in mg equivalents of trolox per 100 g of fresh weight (mg ET·100 g^{-1} p.f.). To calculate the percentage inhibition of the free radical ABTS$^{\bullet+}$, the formula was used: % inhibition = $[(A_0 – A_F) / A_0] \cdot 100$, where: A_0 = initial absorbance of the free radical at 734 nm, A_F = final absorbance of the reaction with the sample.

Results and Discussion

Physical-chemical properties

Significant differences ($P \leq 0.05$) were found between the physicochemical characteristics of the fruits of the four segregants. The P5-3F segregant presented significantly higher fruit weight and size, the smallest and lightest fruits were found in the P5-18F segregant. The same trend was found for seed weight and size (Table 1). Information on the morphological characteristics of the Mexican capulin fruit is limited; however, Hernández Rodríguez et al. (2019) report lower values of weight, polar and equatorial diameters (< 2.8 g, 1.1 cm y 1.2 cm, respectively) of fruits at consumption maturity of *P. Serotina* collected in Zacatlán, Puebla, Mexico, the differences are probably due to their wild nature.

Fruits of segregant P5-1F obtained the highest significant values of total soluble solids (TSS °Brix) and pH, therefore, they were the fruits with the lowest sourness and probably the sweetest. Fruits of segregants P5-3F, P5-18F and P5-28F showed no significant differences in TSS content, while fruits of the segregant P5-3 obtained the lowest pH and TSS values (Table 2). According to Baxter et al. (2005), the increased sucrose discharged from the phloem is the main factor causing differences in soluble solids content (TSS) among fruits of several plants of the same species. This could explain the higher TSS content in the P5-1F segregant. On the other hand, the pH values obtained in this research were higher than those reported by Ordaz-Galindo et al. (1999) (pH 3.96) and Jiménez et al. (2011) (pH 4.20) in fresh pulp of the same species (*Prunus serotina* subsp *capuli)*, this could be due to genetic variability (Ballistreri et al., 2013).

Significant differences ($P \leq 0.05$) were found in the color parameters (*hue* angle of tone, *chroma* and brightnes) of the fruit peel (Table 2). Fruits of segregant P5-1F showed the significantly higher values of *chroma* and *hue* angle of tone (17.16 and 28.35, respectively), which identified them as fruits with higher intensity of orange. Fruits of segregant P5-28F showed the statistically lowest values of *chroma* and *hue* (6.18 and 20.95, respectively), which identified them as redder and lower color intensity fruits. Brightness values allowed dividing the fruits of the four segregants into two groups, the significantly lighter fruits (P5-1F and P5-3F) and the darker fruits (P5-18F and P5-28F). Peel color is the most important attribute of quality and maturity in capulin fruits, associated with the presence of anthocyanins (Hernández Rodríguez et al., 2019; Jimenez et al., 2011).

Mineral content

Significant differences ($P \leq 0.05$) in mineral content were observed among the fresh fruits of the four segregants (Table 4). Segregant P5-1F presented the statistically higher concentration of P, K and Mg, in contrast, fruits of segregant P5-28F showed significantly higher Na, Ca, Fe and Cu contents; segregants P5-1F, P5-28F and P5-3F obtained the highest B contents (Table 3). Luna-Vázquez et al. (2013) reported higher values of K (184.30 ± 3.50 mg·100 g^{-1} p.f.) and Na (22.40 mg·100 g^{-1} p.f.) in capulin fruits harvested in Huejotzingo, Puebla, Mexico; as well as, lower contents of P (28.10 ± 0.40 mg·100 g^{-1} p.f.) and Ca (12.90 ± 1.90 mg·100 g^{-1} p.f); however, the Mg content (21.20 ± 0.40 mg·100 g^{-1} p.f.) was similar to that obtained in the present research. The differences found with respect to the values reported in other investigations are mainly due to edaphoclimatic factors of the place of origin of the capulin harvest. Regarding the differences found in mineral concentrations among the four segregants, they could again be attributed to genetic differences as has been reported in other fruits and some vegetables (Reynoso-Camacho et al., 2006). There are no studies carried out on the variation of mineral content in capulin fruits in relationship to these factors, the present investigation is a contribution of mineral content in segregants. Therefore, the obtained results suggest that the consumption of capulin fruits could be an economical alternative for mineral intake in the population.

Proximal analysis

After fruit processing (40 °C for 5 min), ash content (minerals in the food) decreased only in three segregants P5-1P and P5-28P, as well as humidity content (Table 4); therefore, there was a loss of minerals (leachates) by transfer to the cooking water. According to Yagmur & Taskin (2011), most minerals in fruits and vegetables are water soluble, so it is common for these nutrients to pass from the tissue

Table 2. Total soluble solids content (°Brix), pH and color attributes in fresh fruit of four segregants of capulin (*Prunus serotina*).

Segregant	Total Soluble Solids (°Brix)	pH	Brightness (%)	Angle of tone *Hue* (°)	Chroma
P5-1F	15.88 ± 0.97 a	5.40 ± 0.18 a	23.34 ± 1.46 a	28.35 ± 3.82 a	17.16 ± 2.98 a
P5-3F	14.84 ± 1.43 b	4.21 ± 0.16 d	22.75 ± 1.69 a	19.38 ± 3.95 b	11.64 ± 3.82 b
P5-18F	14.92 ± 0.88 b	4.78 ± 0.16 b	21.22 ± 0.89 b	18.02 ± 2.93 b	9.44 ± 3.17 b
P5-28F	15.37 ± 0.88 ab	4.42 ± 0.20 c	20.95 ± 0.63 b	17.68 ± 2.68 b	6.18 ± 1.91 c

Values reported are the mean of 25 replicates ± standard deviation. Means with the same letter, in the same column are statistically equal (Tukey, 0.05).

Table 3. Mineral content (mg ·100 g $^{-1}$ p.f.) in the fresh fruit pulp of four segregants of capulin (*Prunus serotina*).

	Segregant			
Mineral	P5-1F	P5-3F	P5-18F	P5-28F
P	40.28 ± 0.34 a	35.87 ± 0.24 c	35.85 ± 0.39 c	37.13 ± 0.41 b
K	106.72 ± 0.38 a	100.46 ± 0.46 c	104.99 ± 0.26 b	96.31 ± 0.17 d
Ca	22.63 ± 0.36 b	21.19 ± 0.25 c	16.43 ± 0.32 d	23.82 ± 0.43 a
Mg	24.57 ± 0.07 a	22.34 ± 0.21 b	16.97 ± 0.12 d	19.67 ± 0.42 c
Na	21.03 ± 0.39 ab	20.19 ± 0.42 b	17.45 ± 0.42 c	21.63 ± 0.05 a
Fe	0.67 ± 0.08 b	0.63 ± 0.00 b	0.53 ± 0.00 b	0.88 ± 0.04 a
Mn	0.16 ± 0.00 b	0.17 ± 0.00 b	0.20 ± 0.00 a	0.20 ± 0.01 a
Zn	0.28 ± 0.00 b	0.34 ± 0.00 a	0.30 ± 0.00 b	0.32 ± 0.00 a
Cu	0.02 ± 0.00 d	0.04 ± 0.00 b	0.03 ± 0.00 c	0.07 ± 0.00 a
B	0.99 ± 0.03 a	1.04 ± 0.00 a	0.90 ± 0.01 b	1.01 ± 0.02 a

Values represent the mean of 3 replicates; ± standard deviation. Means with the same letter, in the same row, are statistically equal (Tukey, 0.05).

Table 4. Proximal analysis (%) of the fresh fruits (F) and processed (P) fruits of four segregants of capulin (*Prunus serotina*).

Segregantng	Ashes	Humidity	Protein	Carbohydrates	Crude fiber
P5-1 F	0.771 ± 0.016 d	79.45 ± 0.26 b	1.79 ± 0.46 b	17.95 ± 0.25 ab	3.99 ± 0.06 ab
P 5-3 F	0.954 ± 0.004 a	81.21 ± 0.26 a	2.37 ± 0.01 a	15.52 ± 0.11 d	3.45 ± 0.06 d
P 5-18 F	0.911 ± 0.008 b	80.98 ± 0.66 a	1.82 ± 0.09 b	16.33 ± 0.76 cd	3.63 ± 0.17 cd
P5-28 F	0.912 ± 0.011 b	79.92 ± 0.02 b	1.83 ± 0.01 b	17.31 ± 0.10 bc	3.85 ± 0.01bc
P5-1 P	0.684 ± 0.008 e	79.26 ± 0.11 b	1.82 ± 0.01 b	18.21 ± 0.11 ab	4.04 ± 0.02 ab
P 5-3 P	0.856 ± 0.013 c	81.33 ± 0.09 a	1.17 ± 0.02 d	16.52 ± 0.09 cd	3.67 ± 0.02 cd
P5-18 P	0.841 ± 0.015 c	81.24 ± 0.16 a	1.40 ± 0.02 c	16.42 ± 0.13 cd	3.64 ± 0.03 cd
P5-28 P	0.821 ± 0.011c	78.95 ± 0.16 b	1.47 ± 0.02 c	18.74 ± 0.17 a	4.16 ± 0.04 a

Values represent the mean of 3 replicates; ± standard deviation. Means with the same letter, in the same column, are statistically equal (Tukey, 0.05).

to the process water; the external diffusion of minerals during cooking depends on the level of physical damage to the plant tissues and increases with thermal treatment in the cooking water; factors such as pH level, temperature, water-nutrient ratio, exposed surface area, among other factors affect mineral losses in the final product.

On the other hand, the fresh fruits of the four capulin segregants are an important source of carbohydrates, crude fiber, and protein (Table 4). No significant differences were found in fresh lipid content (F). Lipid content in all segregants was less than 0.03 % and therefore for l it was not reported in Table 5.

The P5-1F fruits had the highest carbohydrate and crude fiber concentrations among the studied segregants. Fruits of segregant P5-3F had the highest crude protein concentration. In this regard, Luna-Vázquez et al. (2013) reported lower carbohydrate values, similar crude fiber and higher protein values in capulin fruits (*P. serotina* subssp. *capuli)* (12.23, 3.58 and 2.10 %, respectively); but the fruit of the studied segregants presented higher values of carbohydrates and protein compared to those reported in cherry (*Prunus domestica)* (8.28 and 0.49 %, respectively) and grape (*Vitis vinifera)* (13.96 and 0.46 %, respectively).

Therefore, this fruit is a source of nutrients at low cost. It is important to highlight that the crude fiber value found in capulin of the studied segregants, between 18.36 to 19.41 % in dry weight, is higher than that reported by Blejan et al. (2023) in some by- products (dry mixture of peels, seeds and residual pulp after juice removal) of wild blueberries (*Vaccinium myrtillus* L.) and blackcurrants (*Ribes nigrum* L.) (11.84 and 15.50 % dry weight, respectively). Foods rich in fiber provide health benefits for the prevention and reduction of the risk of chronic diseases; the consumption of crude fiber has a laxative effect, that is why it is recommended by specialists to people suffering from constipation (Ioniță-Mîndrican et al., 2022). Regarding the effect of thermal treatment on carbohydrate and crude fiber contents, processed fruits (P) showed higher concentrations compared to that of fresh fruit (F) (Table 4), due to the concentration of these nutrients as well as water loss from the fruit during thermal treatment. According to Ramalakshmi et al. (2021), nutrient loss during cooking depends on the temperature, length of treatment and the nutrient involved; carbohydrate loss during cooking is generally small and only after several minutes of cooking and at temperatures close to 100° C.

Finally, it is important to note that a reduction of up to 25 % in protein content was observed in processed fruits (P) with respect to fresh capulin (Table 4), with the exception of the fresh and processed segregant (P5-1F and P5-1P) whose content was statistically equal in both conditions. Considerable loss of soluble nutrients substances when dissolved or leached in the cooking water, such as proteins, water-soluble minerals and vitamins, is common (Deng et al., 2019).

Nutraceuticals content

Significant differences ($P \leq 0.05$) were found in the concentration of phenolic compounds, anthocyanins, flavonoids and vitamin C among the fresh fruits of the four segregants. Fresh fruit (F) of segregants P5-28F and P5-3F showed the highest concentrations of anthocyanins and flavonoid compounds. Regarding the concentrations of total flavonoids and vitamin C, the values were similar among all segregants, except for P5-1 F which presented the lowest concentrations of these metabolites (Table 5). Anttonen & Karjalainen, (2005) report that the content of phenolic components can vary significantly among cultivars of a species due to gene expression related to the biosynthesis of some metabolites in response to changes in the crop environment. Phenolic components in addition to their antioxidant capacity possess other mechanisms of action that explain their diverse beneficial effects on consumers (Potì et al., 2019).

Phytochemical studies of cultivars, varieties or segregants of a species allow planning breeding strategies, as

Chart 5. Content of nutraceutical compounds in fresh fruit (F) and processed (P) of four capulin segregants (*Prunus serotina)* by each 100 g of fresh weight.

Segregant	Phenolic Compounds (mg EAG)	Flavonoids (mg EQ)	Anthocyanins (mg ECyd-3-Gli)	Vitamin C (mg EAA)
P5-1 F	96.42 ± 3.09 e	28.39 ± 0.18 e	9.05 ± 0.20 e	33.87 ± 0.24 c
P5-3 F	331.57 ± 4.09 b	50.49 ± 0.83 a	19.69 ± 0.19ª	40.06 ± 0.55 a
P5-18 F	228.84 ± 5.95 d	48.68 ± 1.47 ab	16.46 ± 0.61 c	42.01 ± 0.93 a
P5-28 F	341.27 ± 3.09 b	50.25 ± 0.44 a	18.54 ± 0.32 ab	40.46 ± 0.77 a
P5-1 P	104.6 ± 2.14 e	31.30 ± 0.31d	8.44 ± 0.24 e	33.52 ± 0.34 c
P5-3 P	390.69 ± 3.24 a	49.58 ± 0.53 a	16.57 ± 0.34 c	40.06 ± 0.62 a
P5-18 P	305.27 ± 2.14 c	46.44 ± 0.53 b	10.77 ± 0.24 d	36.61 ± 0.40 b
P5-28 P	388.84 ± 0.86 a	42.01 ± 0.12 c	17.48 ± 0.29 bc	36.69 ± 0.54 b

Values represent the mean of 3 replicates; ± standard deviation. Means with the same letter, in the same column, are statistically equal (Tukey, 0.05). EAG: gallic acid equivalents, EQ: quercetin equivalents, ECyd-3-Gli: cyanidin-3-glucoside, EAA: ascorbic acid equivalents.

well as selecting individuals with high content of active ingredients or commercial interest as natural colorants, nutraceutical ingredients, and antioxidants for the food industry or to improve the content of healthy compounds in capulin fruits.

It is important to note that the values of phenolic compounds and flavonoids (4.69-17.64 mg EAG $\cdot g^{-1}$ y 1.38-2.68 mg EQ$\cdot g^{-1}$ p.s., respectively) found in the four studied segregants and transformed in the same concentration units were lower than those reported by Hernández Rodríguez et al. (2019) in wild capulin collected in Zacatlán, Puebla, Mexico (14.40-26.96 mg EAG$\cdot g^{-1}$ p.s., 16.56-9.23 mg EQ$\cdot g^{-1}$ p.s. and 0.04-0.66 mg cyanidin-3-glucoside(C-3-G)$\cdot g^{-1}$ p.s.); in contrast, higher anthocyanin concentrations (0.44-1.05 mg C-3-G equivalents per g, dry weight) were found in all the studied segregants. Ordaz-Galindo et al. (1999) reported anthocyanin values in capulin fruits (*Prunus serotina*) (31.7 mg equivalents of cyanidin -3-glycoside$\cdot$100 g^{-1} p.f.) similar to those reported in the present research.

On the other hand, the concentration of flavonoids found in all the segregants was lower than that of total phenolic components, possibly because some flavonoids could be found forming procyanidins (condensed tannins) as in other fruits (Cui et al., 2006).

The vitamin C content was statistically equal among the fresh fruits of segregants P5-3F, P5-18F and P5-28F, the lowest value of this metabolite was present in the fresh fruits of segregants P5-1F. There are no studies on the content of this vitamin in capulin.

On the other hand, thermal treatment had a significant effect on the content of nutraceutical compounds among the fruits of the four segregants. The content of phenolic components obtained a significant increase in the fruits of the four segregants post- thermal treatment (17 %), which could be due to the concentration of these nutrients by the loss of water from the fruit during thermal treatment, as was observed in carbohydrates.

Segregants P5-3F and P5-18F showed no significant differences in flavonoid content after thermal treatment, while segregant P5-28P showed the greatest decrease in these metabolites. Flavonoids are also phenolic compounds with antioxidant potential found in vegetables and fruit; in the latest two decades epidemiological studies have shown a relationship between flavonoid consumption and the low incidence of degenerative diseases (Toh et al., 2013). The mechanisms of action of each group of phytochemicals in capulin are unknown, but the synergistic effect of these bioactives make it a food with notable functional properties, mainly the fruits of the segregant P5-3F and P5-28F.

The effect of the thermal treatment also resulted in a significant average decrease of 16 % of anthocyanin content in P5-3F and P5-18F segregants (Table 5). Anthocyanin levels may be affected by the temperature of process. Oliveira et al. (2010) observed a reduction in anthocyanin content in blueberries between 12 and 42 % during progressive heating from 12° to 99 °C for 60 min, the same phenomenon was found in the thermal treated capulin studied in the present research.

Vitamin C content (Table 6) showed significant differences only between fresh and processed fruits of segregants P5-18F and P5-28F, where a decrease in content was generated (12.8 and 9.3 %, respectively). The vitamin C content in fresh capulin fruits of the four analyzed segregants was higher than that reported by Garcia et al. (2006) in banana (8 – 16 mg$\cdot$100 g^{-1}) and green apple (3-30 mg$\cdot$100 g^{-1}). Vitamin C plays a very important role in human metabolism, it is essential for the development and function of the nervous system, it is part of the mechanisms of cicatrization, biosynthesis of collagen and different neurotransmitters (Kükürt & Gelen, 2024).

Table 6. Antioxidant capacity in fresh (F) and processed fruits (P) of four segregants of capulin (*Prunus serotina*).

Segregant	Antioxidant Capacity (µ mol· ET100 g^{-1} pf)	Inhibition (%)
P5-1 F	1326.084 ± 47.27 c	54.17 ± 2.17 b
P5-3 F	1800.81 ± 23.23 b	92.25 ± 3.28 a
P5-18 F	1390.40 ± 2.88 c	57.13 ± 0.13 b
P5-28 F	2154.05 ± 71.48 a	95.77 ± 1.13 a
P5-1 P	1816.94 ± 8.81 b	91.36 ± 0.40 a
P5-3 P	2252.06 ± 22.22 a	96.76 ± 1.02 a
P5-18 P	2145.89 ± 76.11 a	91.87 ± 3.50 a
P5-28 P	2134.66 ± 8.78 a	97.84 ± 0.43 a

Values represent the mean of 3 replicates; ± standard deviation. Means with the same letter, in the same column, are statistically equal (Tukey, 0.05). ET: trolox equivalents.

Finally, it is important to point that the fresh fruits of the P5-3F segregant were the ones in which nutraceutical compounds were less affected to the thermal treatment (except for the concentration of anthocyanins), which corresponded with higher antioxidant activity (Table 6), for this reason, it is recommended as a segregant with potential to be processed.

Antioxidant capacity

The antioxidant capacity of the fresh and processed fruits of the four segregants showed significant differences ($P \leq 0.05$) (Table 6). The highest antioxidant capacity values were observed in the fresh fruit of segregant P5-28F, followed by P5-3F. The fresh fruits of these segregants had the highest content of anthocyanidins, phenolic compounds, flavonoids and vitamin C. The processed fruits of the four segregants showed an increase of 28 % in the antioxidant capacity content; as well, in their average of free radical inhibitory capacity (34 %), which could be explained by the increase of phenolic compounds (17 %) as mentioned above in the present study. The antioxidant capacity obtained in fresh and processed fruits of the four capulin segregants (1326.08 ± 47.27 – 2252.06 ± 22.22 µmol ET·100 g^{-1} p.f.) was similar to the values reported by Luna-Vázquez et al. (2013) (1455.2 ± 92.5 – 2056.7 ±108.0 µmol ET·100 g^{-1} p.f.) in fresh fruits of the same species. On the other hand, similar research carried by García-Mateos et al. (2013) on 20 different tejocote genotypes and Ballistreri et al. (2013) on 24 sweet cherries (*P. avium*), varieties showed that genetic factor could explain variations in the nutraceutical characteristics of capulin segregants.

Conclusions

The capulin fruit of the four studied segregants is a source of nutrients (protein, fiber, carbohydrates, P, K, Ca, Mg and Fe) and antioxidant compounds (phenolics, anthocyanins and vitamin C) at a low cost. The fruit of segregant P5-1F had the highest carbohydrate content, total soluble solids and pH, important quality attributes for marketing a fruit; however, it had the lowest nutraceutical value in the fresh and processed condition. In contrast, the fruits of P5-3 segregant had the highest protein values and nutraceutical potential due to their high concentrations of phenolic compounds, flavonoids, anthocyanins, vitamin C and antioxidant activity in its fresh (P5-3F) and processed (P5-3P). Fresh fruits from segregants P5-3F and P5-28F were statistically superior in anthocyanin and flavonoid content, sharing this superiority in vitamin C content with fresh fruits from segregant P5-18F. The fresh fruits of the P5-28F showed the highest antioxidant capacity, associated with their high concentration of nutraceutical compounds. The factors thermal treatment (40 °C for 5 min) and capulin segregant had a joint significant effect on the nutritional and nutraceutical value of fresh and processed fruits. The fruits of P5-3P segregant tolerated the thermal treatment the most, so the fruits showed the least effects in the nutraceutical components and the highest value of antioxidant activity, for this reason it is recommended as a segregant with potential to be processed in order to generate added value.

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Dendrochronology in Mexico over the last seventy years

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Abstract

The study of dendrochronology has intensified in recent years due to its close relationship with addressing environmental issues. Therefore, the objective of this study was to analyze the development of dendrochronological research in Mexico through a comprehensive literature review, which included search engines, websites, and publications from national and international journals. The collected information was organized into a database and classified into nine categories to analyze study frequencies, species, and locations. A total of 229 documents published between 1944 and 2021 were identified, with a significant increase during the period from 2009 to 2021, when dendroclimatology emerged as the most studied subdiscipline. In total, 429 different chronologies were documented, primarily distributed in the Sierra Madre Occidental. The best-represented reconstructed period spanned from 1801 to 2019. The most frequently studied species were *Pseudotsuga menziesii* and *Pinus hartwegii*. The most common application of this science has been precipitation reconstruction. Due to its extensive biodiversity, Mexico has significant potential for dendrochronological research. However, it is essential to expand both the physical and intellectual boundaries of this field and broaden its spatial scope within the country.

Keywords: tree rings, chronologies, dendroclimatology, dendroecology, climate reconstruction.

Introduction

The development of forest ecosystems is primarily determined by environmental factors. When these factors align with optimal conditions, tree growth improves; conversely, under adverse conditions, growth tends to be limited (Villanueva-Díaz et al., 2011). The radial growth of woody forest species, particularly conifers, leads to the formation of growth rings. These anatomical structures inherently record temporal variability and influence of factors such as climate, wildfires, pests, diseases, geomorphological processes, competition, air pollution, and human management, among others (Franco-Ramos & Vázquez-Selem, 2017). Among these factors, climate has the greatest influence on natural forests (Rojas-García et al., 2020).

Dendrochronology is a science that examines temporal variability of physical, structural, and compositional changes in tree growth rings and their relationship with environmental conditions of the sites where they develop. This discipline makes it possible to reconstruct and study a part of the environmental history underlying the development of the

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trees, as it allows for the analysis of various climatic variables and ecological processes spanning from hundreds to thousands of years (Douglas, 1941).

Dating growth rings of species sensitive to specific environmental conditions has become a trusted tool for the historical analysis of these conditions, as it offers a wider temporal perspective than instrumental records (Villanueva-Díaz et al., 2002). Depending on the approach and application of dendrochronological series, this science is categorized into various fields, including dendroclimatology, dendroecology, dendropyrology, dendrochemistry, dendrogeomorphology, dendrohydrology, dendrovolcanology, and dendroarchaeology (Amoroso & Suarez, 2015).

The study of dendrochronology in Mexico began in the 1940s with the work of Schulman (1944). In the early 1960s, new chronologies were established during the expedition to Casas Grandes in Chihuahua, where a notable 500-year floating chronology was developed (Bannister & Scott, 1964). In the 1970s, the "Mexican Tree Ring Project" led to the development of several chronologies (Therrell, 2003), although their application has remained limited (Villanueva-Díaz et al., 2011). During this time, Naylor (1971) conducted a preliminary study in Oaxaca, but was unable to generate successful chronological advances.

At the beginning of the 21st century, the global interest in understanding the historical variability of climate and other environmental factors influencing forest dynamics (such as wildfires and atmospheric circulation phenomena) spurred the study of dendrochronology in Mexico. This was achieved through technical and financial contributions of international organizations (Villanueva-Díaz et al., 2002). In the last two decades, the use of multiple forest species with dendrochronological potential across various approaches has demonstrated Mexico's viability for the development of this science (Reyes-Basilio et al., 2020). These advancements have positioned the country as one of the leading nations in dendrochronological research in Latin America (Fo et al., 2009).

In this context, the objective of this article was to analyze the spatial-temporal evolution of dendrochronology studies in Mexico through a comprehensive literature review and the analysis of relevant scientific products. The goal was to generate information that provides a comprehensive understanding about the application of this science in the country, to diagnose research needs and opportunities, and to provide solid bases for decision-making for the development of dendrochronology.

Materials and methods

For this study, scientific publications (research articles) as well as gray literature (books and chapters, conference proceedings, technical brochures, and preprints) from 1944 to 2021 were considered. The latter, although less formal, represent relevant contributions from the scientific community that, directly and indirectly, are related to the progress of dendrochronology in Mexico (Montes de Oca-Montano, 2018). Information was searched and obtained through specialized search engines such as Science Direct, Scopus, Wiley Online Library, and Springer; additionally, websites like Google Scholar, Redalyc, SciELO, and PubMed were consulted. Special attention was paid to the contents of the main national forestry journals: Revista Mexicana de Ciencias Forestales, Madera y Bosques, Revista Chapingo Serie Ciencias Forestales y del Ambiente, Agrociencia, and Investigaciones Geográficas, as well as high-impact international journals: Dendrochronologia, Tree-Ring Research, Trees y Forest.

For the search, the following keywords were used in both Spanish and English: "dendrochronology", "growth rings", "Mexico", "dendroclimatology", "dendroecology", "radial growth", "chronologies", "climatic reconstruction", "tree rings", "fire scars" y "paleoclimatology", as well as their combinations using the logical operator AND. As inclusion criteria, all publications referring to dendrochronology in Mexico were considered.

The information collected was organized into a database in Microsoft Excel®, using macros to semi-automate the process. The information fields corresponded to two criteria: documents and content. The first criterion included a unique identifier composed of consecutive numbers, title, year of publication, journal, authors and institution of origin. For the content of the documents, the following was considered: study site (entity, latitude, longitude and altitude), species studied, type of vegetation, variable measured in the rings (ring width, earlywood, latewood, chemical composition, fire scars or geomorphic processes, among others), chronology ranga and period, reconstructed variable, and range and period of reconstruction.

Subsequently, the information was classified into nine categories based on the analysis of growth ring chronologies: dendroclimatology, dendroecology, dendropyrology, dendrochemistry, dendrogeomorphology, dendrohydrology, dendrovolcanology, dendroarchaeology, and others (which included systematic reviews, meta-analyses, sites suitable for dendrochronological studies, and reconstruction of agro-food variables). Each category was analyzed based on frequencies, species, and locations studied.

Results and discussion

A total of 229 documents related to dendrochronology in Mexico were identified. Of these, 90 % (206) were scientific articles, while books or book chapters accounted for 6.6 % (15), proceedings from scientific dissemination events made up 3 % (seven), and preprints represented 0.4 % (one).

The scientific articles were published in a wide range of indexed journals. A total of 83 different journals were identified, mainly foreign (74.4 %). This reflects the quality and scientific rigor of research in this field in Mexico, because these articles met the requirements for publishing their results in leading journals. More than half (53.66 %) of the published articles appeared in foreign journals, primarily from the United States (Figure 1). The national journals with the highest number of publications were Revista Mexicana de Ciencias Forestales, Madera y Bosques, and Revista Chapingo Serie Ciencias Forestales y del Ambiente.

Between 1944 and 2021, scientific production on dendrochronology in Mexico concentrated in 33 years. The average production during this period was 2.66 articles per year (205 articles in total). Although the first studies were conducted in the 1940s (Schulman, 1944), the initial uncertainty about Mexico's potential for the development of this discipline led to intermittent progress and the generation of a limited number of chronologies throughout the 20th century (Bannister & Scott, 1964; Villanueva-Díaz et al., 2000; Naylor, 1971). In contrast, the 21st century saw a significant shift in the research trend of this science.

Three periods of scientific production in dendrochronology in Mexico were identified (Figure 2). The first period (from 1944 to 1998) showed limited development and low productivity in dendrochronological research in the country. During these 54 years, only 12 scientific products were recorded (5 % of the total). According to Villanueva-Díaz et al. (2000), just over 40 chronologies were generated during this period.

In the second period (from 1999 to 2008), a substantial improvement was observed, with 17 % (39) of the 229 scientific products found published. Villanueva-Díaz et al. (2009) classify this period as one in which the knowledge of the application of dendrochronology in Mexico was strengthened. This resurgence was driven by four factors: 1) growing scientific interest in climate elements and their temporal variation, atmospheric circulation phenomena (Villanueva-Díaz et al., 2002), temporal availability of water resources (Villanueva-Díaz et al., 2007), and influence of other environmental factors on natural resources; 2) the establishment of a dendrochronology laboratory in Mexico; 3) national and international funding for research projects (Villanueva-Díaz et al., 2008); and 4) collaboration and technical support of international organizations (Villanueva-Díaz et al., 2011).

In the third period (from 2009 to 2021), the rise of dendrochronology in Mexico was consolidated, with 77.7 % (178) of the scientific products considered in this study being published.

The temporal analysis identified nine categories or research approaches, with the results emphasizing the predominance of studies focused on the relationship

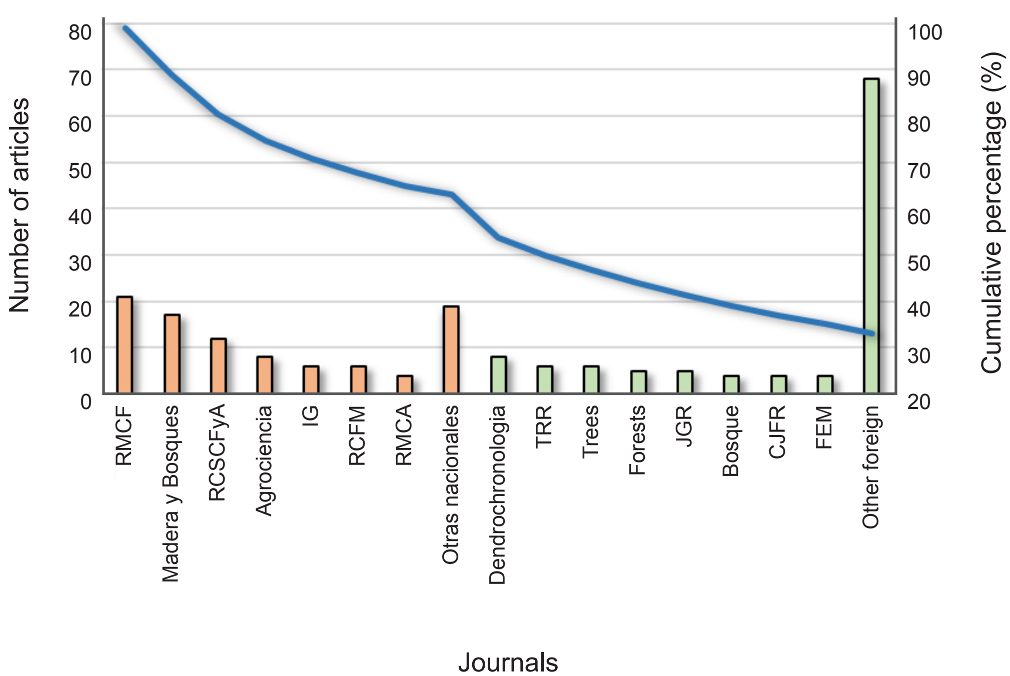

Figure 1. Distribution of the number of articles and national (orange) and foreign (green) journals that have published research on dendrochronology in Mexico. The blue line reflects the cumulative percentage. RMCF: Revista Mexicana de Ciencias Forestales; RCSCFyA: Revista Chapingo Serie Ciencias Forestales y del Ambiente; IG: Investigaciones Geográficas; RCFM: Revista de Ciencias Forestales en México; RMCA: Revista Mexicana de Ciencias Agrícolas; TRR: Tree-Ring Research; JGR: Journal of Geophysical Research; CJFR: Canadian Journal of Forest Research; FEM: Forest Ecology and Management.

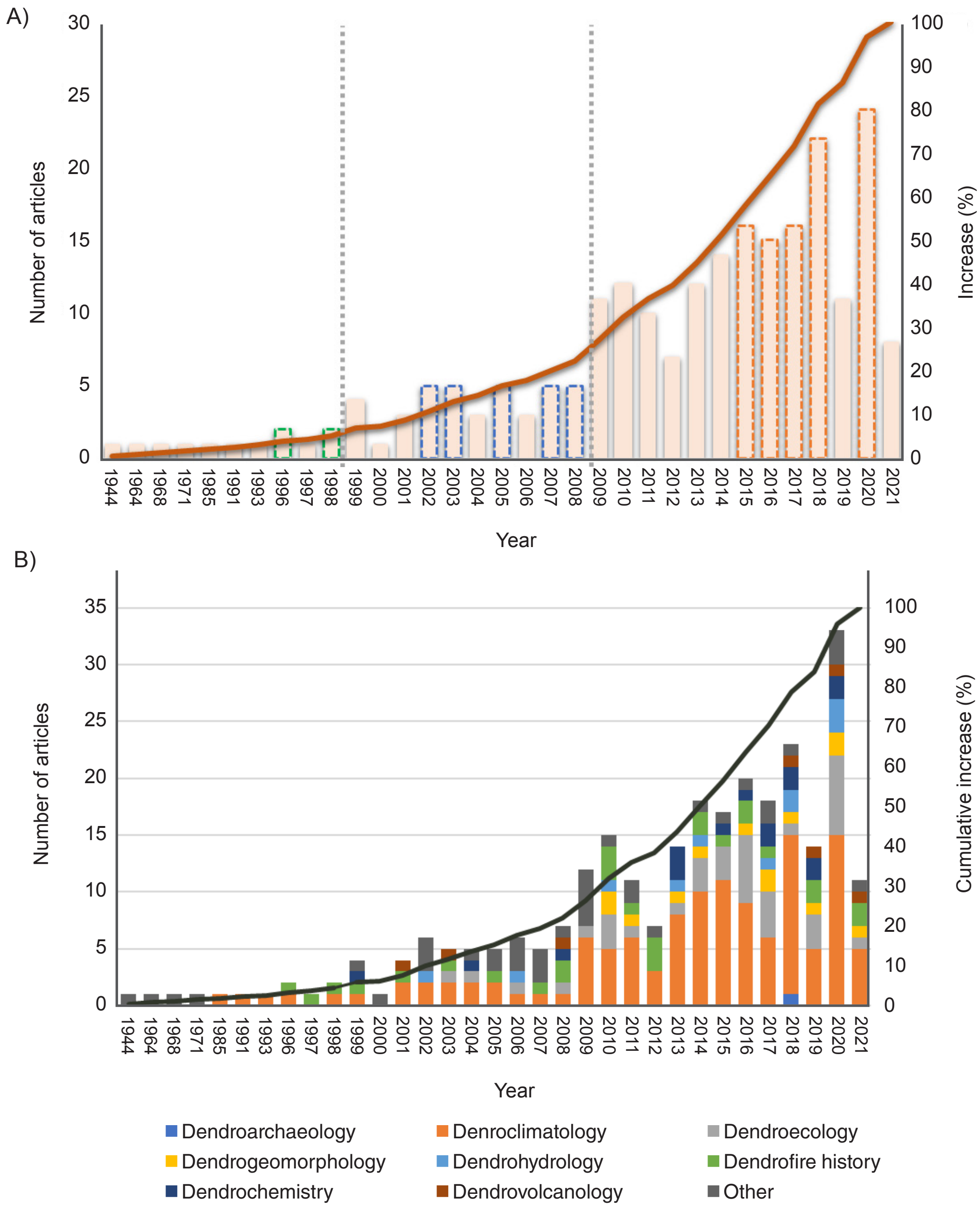

Figure 2. Scientific production related to dendrochronology in Mexico: A) three periods of scientific production (highlighted bars indicate outstanding production) and B) production according to the dendrochronology category.

between ring width and climate variables. Other approaches include research on the influence of environmental variables other than climate (Grissino-Mayer et al., 2005), dendrochronological potential, dissemination of methodologies, and innovation studies (Figure 2B).

Dendroclimatology

Dendroclimatology is based on the physiological ability of certain tree species to record climatic variability in their growth rings, making them genuine meteorological biostations (Cerano-Paredes et al., 2016, 2021). This physiological ability allows them to record dominant climate conditions, as well as interannual variations at a specific spatial scale and with seasonal resolution (Manzanilla-Quiñones et al., 2020). Dendrochronological methods help detect the relationship between ring width and climate variables, leading to the emergence of dendroclimatology. This subdiscipline is essential for analyzing global climate variability, the influence of atmospheric circulation phenomena, and their effects on natural resources (Cerano-Paredes et al., 2011; Gómez-Guerrero et al., 2013).

In Mexico, dendroclimatology represents the most developed approach within this science, with 53 % (122) of the scientific products analyzed. Research is mainly concentrated in the north and center of the country and has made it possible to evaluate how climatic variables influence forest development in these regions. In addition, they have made possible the reconstruction of climatic variables, such as seasonal precipitation and seasonal rainfall. (Cardoza-Martínez et al., 2018; Cerano-Paredes et al., 2011, 2021; Chávez-Gándara et al., 2017; Díaz-Ramírez et al., 2016; Irby et al., 2013; Manzanilla-Quiñones et al., 2018; Villanueva-Díaz et al., 2007, 2008, 2009).

Other climatic variables, such as temperature (Villanueva-Díaz et al., 2020) and evaporation (Pompa-García et al., 2013), have also shown a relationship with the development of growth rings. This connection has facilitated the identification of extreme events, such as drought occurrences (Martínez-Sifuentes et al., 2019).

The influence of atmospheric circulation patterns on the climatic variability of Mexico has generated significant interest (Pompa-García et al., 2015). These dendroclimatic studies have enabled the analysis of phenomena such as El Niño-Southern Oscillation (ENSO) (Cerano-Paredes et al., 2011; Pompa-García et al., 2013, 2015). Additionally, the relationship between adverse events and climate variability has been explored (Burns et al., 2014). Overall, the application of dendroclimatology has triggered the creation of an extensive network of chronologies and has allowed the evaluation of dendrochronological potential of a wide range of species (Acosta-Hernández et al., 2020).

Dendroecology

Trees can record certain ecological processes, and dendroecology, as a specialized branch of dendrochronology, focuses on their study (Rojas-García et al., 2020). This discipline has been used to identify the temporal occurrence of natural processes that influence the development of growth rings, making it possible to determine the significance of these processes for the development of forest ecosystems. In Mexico, dendroecology has had several applications. By analyzing stand structures and applying dendrochronology, researchers have estimated forest ages (Villanueva-Díaz et al., 2010), biomass quantities (Correa-Díaz et al., 2019, 2020; Martínez-Sifuentes et al., 2019), forest productivity (Arreola-Ortiz & Návar-Cháidez, 2010; Castruita-Esparza et al., 2016; Gómez-Guerrero et al., 2015; Reyes-Cortés et al., 2020) and the capacity of forests to capture carbon (García-Bedolla et al., 2015; Reyes-Basilio et al., 2020).

Other applications include evaluating the effects of extreme climatic events on the development of natural resources (Acosta-Hernández et al., 2020; Pacheco et al., 2020; Pompa-García et al., 2017; Rodríguez-Ramírez et al., 2018), assessing the impact of pests on tree radial growth (López-Sánchez et al., 2017), and analyzing wood anatomy and density (Morgado-González et al., 2019; Rodríguez-Ramírez et al., 2020).

Dendropyrochronology

Fire plays a crucial ecological role in certain forest ecosystems, and its benefits have been recognized by the scientific community. Dendropyrochronology provides tools for dating wildfires by analyzing the scars left by fire in tree growth rings. This allows for the estimation of parameters related to historical fire regimes (Cerano-Paredes et al., 2019) and provides a solid foundation for detecting anthropogenic disturbances, as well as for developing fire management strategies in forests (Sáenz-Ceja & Pérez-Salicrup, 2019).

In Mexico, this subdiscipline is becoming increasingly common, although studies remain scarce compared to the country's vast forested area (Cerano-Paredes et al., 2021; Sáenz-Ceja & Pérez-Salicrup, 2019). Most studies have identified changes in fire regimes, highlighting periods of fire exclusion, particularly in recent decades (Cerano-Paredes et al., 2021; Fulé & Covington, 1999; Sáenz-Ceja & Pérez-Salicrup, 2019; Yocom et al., 2014). These changes, mainly attributed to human activity, have raised concerns within the scientific community, as prolonged fire suppression can increase the risk of more severe future events due to the accumulation of combustible materials. Moreover, fire is an integral part of the ecology of many forests (Cerano-Paredes et al., 2021). Several studies have recommended incorporating fire management practices, such as prescribed burns, to reduce fuel loads and mitigate the risk of severe wildfires (Cerano-Paredes et al., 2021; Ponce-Calderón et al., 2021).

Some studies, such as that of Skinner et al. (2008) have explored the potential correlation between climate and fire occurrence. However, Fulé and Covington (1999) argue that this relationship is weak. These differences may be attributed to the specific conditions of each study site, including geographic location, topography, and vegetation type. Additionally, Stephens et al. (2010) analyzed the formation of fire scars and found that only a small proportion of trees exhibited scarring.

Dendrochemistry

Trees can absorb chemical components present in water, soil, and air. Variations in pollution levels or in the chemical composition of any of these media can be reflected in the chemical content of growth rings (Reyes-Camarillo et al., 2020). Through dendrochronological analysis, it is possible to determine the temporal variability of the chemical content in tree rings. Dendrochemistry is the subdiscipline that, with the support of chemical methods, allows for the evaluation of trees' physiological responses to variations in chemical elements (Correa-Díaz et al., 2020; Gómez-Guerrero et al., 2013).

In Mexico, dendrochemistry has received little attention in recent years. Studies have focused on three main areas: 1) analysis of stable isotopes and tree physiology (Beramendi-Orosco et al., 2018; Correa-Díaz et al., 2020; Gómez-Guerrero et al., 2013; Pacheco et al., 2020), 2) the impact of environmental pollution on chemical concentration in tree rings (Flores et al., 2017; Morton-Bermea et al., 2016), and 3) the determination of chemical element composition (Sheppard et al., 2008).

Dendrogeomorphology

In mountainous areas, characterized by steep slopes, massive movements of rocky and edaphic material are common, favored by gravity and geological, climatic and anthropic factors. These processes can cause damage and the removal of forest masses. Rock falls, landslides and debris flows can cause alterations in trees, which can be expressed as scarring, abrupt growth reduction, eccentric growth and traumatic resin ducts. Each alteration can be dated through the analysis of growth rings, which facilitates the reconstruction of past geomorphological processes (Stoffel et al., 2011).

In the mountains of Mexico, especially in the Sierra Transversal, debris flows, torrential floods (Franco-Ramos et al., 2019; Martínez-Sifuentes et al., 2019) and lahars (Bollschweiler et al., 2010; Franco-Ramos et al., 2016) have been reconstructed. Additionally, determining the age of trees inhabiting newly formed geomorphological surfaces has allowed estimating the minimum age of these structures (Franco-Ramos & Vázquez-Selem, 2017).

Dendrohydrology

Dendrohydrology is used as a reliable tool for reconstructing river flows, groundwater levels, lake level changes, and floods. By analyzing growth rings, it is possible to generate dendrohydrological reconstructions that provide insights into water resource availability (Villanueva-Díaz et al., 2018). In Mexico, this subdiscipline has been applied to reconstruct streamflow volumes, water body gauge levels, and runoff patterns (Therrell et al., 2006; Villanueva-Díaz et al., 2020).

Dendrovolcanology

The emission of ash and gases from volcanic eruptions can cause alterations in regional environmental conditions. When trees are partially or completely covered by volcanic ash, they may either perish or record abrupt changes in their development, such as the suppression of ring growth (Biondi et al., 2003).

Dendrochronological analysis allows for the reconstruction of eruptive events and the assessment of their effects on forest ecosystems (Torbenson, 2015) by linking volcanic eruptions with tree rings. However, for dendrovolcanological studies, it is crucial to distinguish between climate-induced stress signals and those caused by volcanic activity (Biondi et al., 2003).

In Mexico, dendrovolcanology has gained importance through the analysis of the effects of the volcanoes along the Trans-Mexican Volcanic Belt on trees in the region. These studies have enabled the reconstruction of eruptive events (Alcalá-Reygosa et al., 2018; Alfaro-Sánchez et al., 2020; Sheppard et al., 2008) and the tracing of pyroclastic flows (Franco-Ramos et al., 2019). Furthermore, it has been shown that volcanic eruptions lead to changes in the chemical composition of growth rings, which is why these studies are often complemented with dendrochemical analyses (Alfaro-Sánchez et al., 2020; Carlón-Allende et al., 2015).

Dendroarchaeology

In the past, wood played a crucial role in construction, especially from species that form growth rings. This characteristic has allowed dendrochronology to be used at archaeological sites with ancient wood, for dating historical buildings and other archaeological applications (Bernabei & Macchioni, 2012). While this tool is useful for extending chronologies and inferring historical events, the application of dendroarchaeology in Mexico has seen limited success. Villanueva-Díaz et al. (2011) note that until the early 2010s, there was only one successful example of this subdiscipline applied to prehistoric ruins in Casas Grandes, Chihuahua. Additionally, with the help of the “Wiggle Matching” radiocarbon, it was possible to date ancient wood obtained from two sites in the Malpaso Valley, Zacatecas (Turkon et al., 2018).

Other categories

The studies include those focused on systematic reviews and meta-analyses specific to Mexico (Acosta-Hernández et al., 2017; Villanueva-Díaz et al., 2000), as well as reviews on the development of dendrochronology in a broader region (Bannister & Scott, 1964; Giraldo-Jiménez, 2011; Rojas-García et al., 2020; Schulman, 1944). Additionally, suitable sites for dendrochronological studies (Carlón-Allende et al., 2015) and for the reconstruction of agro-food variables (Therrell et al., 2006) have been identified.

Figure 3 shows spatial distribution and volume of studies related to different categories of dendrochronology. Most of the research has been conducted in the central and northern parts of the country, with a higher concentration in the latter due to the predominance of dendroclimatic studies, both in terms of volume and spatial applicability.

A total of 429 chronologies were identified, primarily distributed across three physiographic provinces: the Sierra

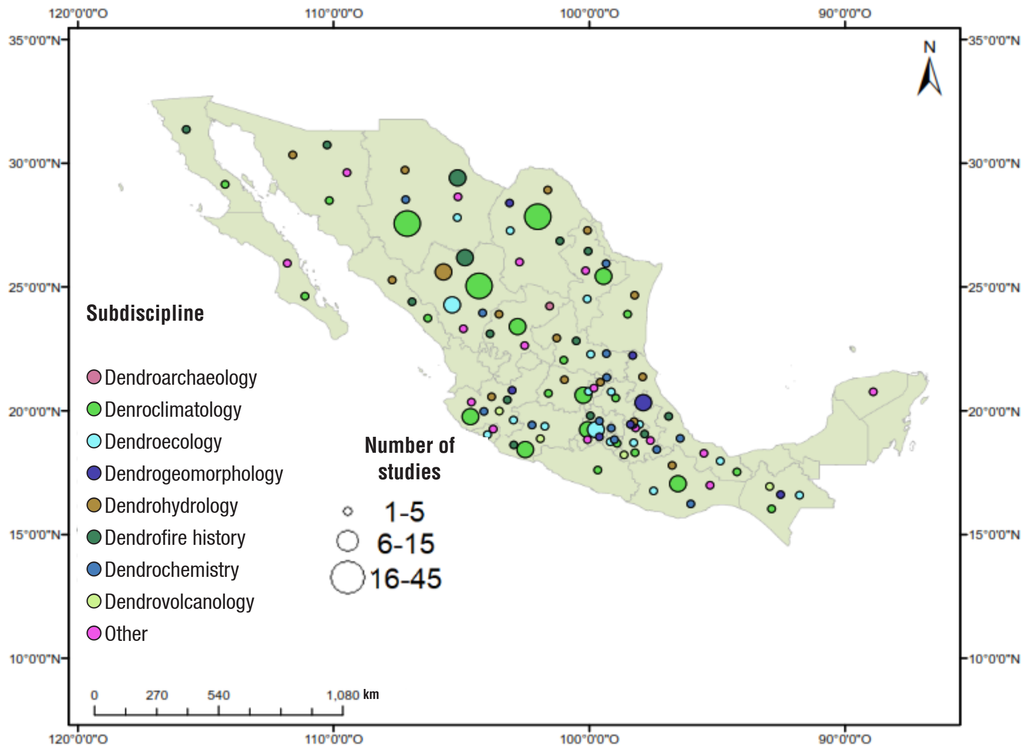

Figure 3. Spatial distribution of dendrochronology subdisciplines in Mexico.

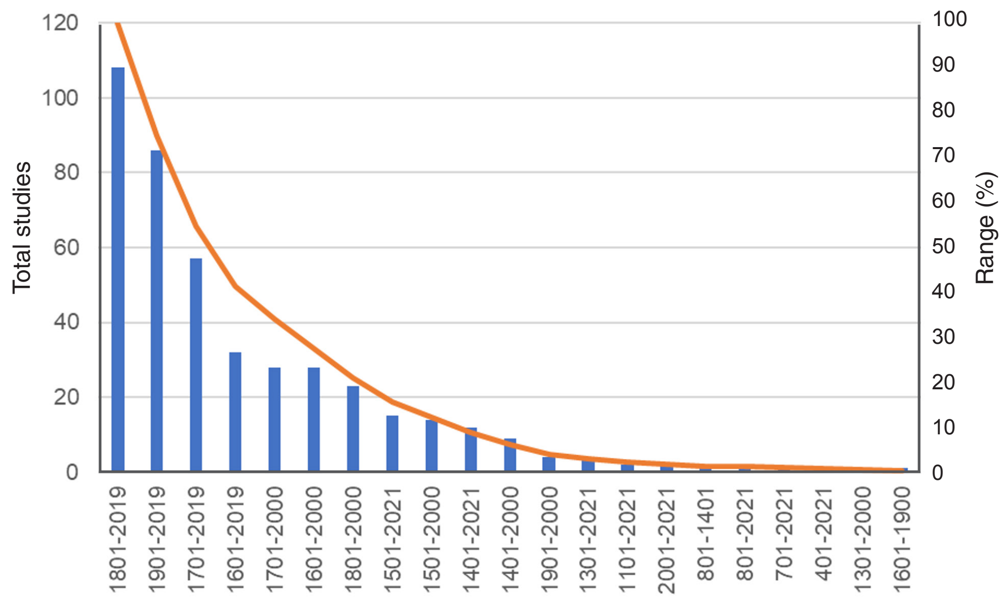

Figure 4. Range (period of years) of tree ring chronologies in Mexico

Madre Occidental (44.76 %), the Trans-Mexican Volcanic Belt (22.38 %), and the Sierra Madre Oriental (19.58 %). The remaining 13.28% is distributed across other provinces in the country. Chronologies have been developed in 28 states of Mexico, with Durango, Chihuahua, and Coahuila accounting for just over half (51 %). Regarding the species used, 53 different species have been reported, with the majority belonging to the conifer group (84.9 %), specifically the genus *Pinus* (60.38 %). Only eight broadleaf species have been used to create chronologies in Mexico, and three species dominate in terms of use: *Pseudotsuga menziesii* (23.78 %), *Pinus hartwegii* (10.26 %) and *Taxodium mucronatum* (8.86 %).

The chronologies span from the year 467 to 2019, covering a period of 1 537 years. The longest chronology was created from the growth rings of *Taxodium mucronatum* in Los Peroles, San Luis Potosí (Villanueva-Díaz et al., 2007). However, since most of the chronologies are concentrated between 1801 and 2019, this period is the best represented (Figure 4). In Mexico, precipitation is the variable most closely related to the variability of tree rings, making it the most reconstructed variable.

Conclusions

Dendrochronological studies in Mexico have experienced notable growth since 1999, successfully exploring various approaches. However, there remains a dominance of climate variables as the primary factors influencing the variability in tree ring widths. Other research approaches in dendrochronology present opportunities for new studies in other regions of the country, particularly in the south.

Broadleaf species with dendrochronological potential need to be studied more in-depth, especially those found in tropical environments, where climatic seasonality is less pronounced. The vast biodiversity of Mexico's forest ecosystems offers an opportunity to expand knowledge in this area.

Dendrochronology in Mexico has significant potential for development. Future studies could focus on comparing the number of studies and species analyzed at an international level. It is essential to broaden both the physical and intellectual boundaries, as well as expand the spatial scope of dendrochronological research in the country.

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Establishment and Monitoring of a *Pinus greggii* Engelm. ex Parl. Plantation as a Climate Change Mitigation Strategy in Huehuetla, Hidalgo

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Abstract

Climate change, as a global phenomenon, has affected the planet in multiple ways, altering weather patterns and prompting the search for alternatives and strategies for mitigation and adaptation. Forest plantations have emerged as an environmental option due to their potential for CO_2 capture, making it essential to identify tree species that can contribute to this effort. One method for evaluating a species' adaptability to specific climatic conditions is climatic matching. In this study, *Pinus greggii* Engelm. ex Parl. was evaluated for its potential to adapt to the subtropical conditions of Huehuetla, Hidalgo—a region with high potential for restoring degraded lands and supporting fast-growing species. Climatic parameters considered included cardinal temperatures, precipitation range, and altitude, along with tree measurement characteristics: height and basal diameter. The species showed a 50 % adaptation rate to the natural conditions, primarily attributed to altitude, given a 300-meter difference between the planting site and the lower distribution limit of the species. Growth was deemed acceptable, as after 24 months of monitoring and evaluation, the trees reached an average height of 94.6 cm and a basal diameter of 1.6 cm. Individual trees exhibited a diameter at breast height of 1.14 cm, which translated to an average volume of 147.5 cm^3 per tree, equivalent to 61.81 g of stored carbon.

Keywords: Climate homologation, *Pinus greggii*, Engelm. Ex Parl., environmental services.

Introduction

In recent years, forest plantations in Mexico have increased due to the opportunity they represent for enhancing timber production. These plantations are defined as the establishment and management of species on land designated primarily for forest use. As a result, the total area covered by commercial forest plantations (CFP) nationwide has reached 373 127.17 ha, of which timber species account for 85.2%. These plantations are primarily distributed across the states of Veracruz, Tabasco, and Campeche (CONAFOR, 2024; Diario Oficial de la Federación, 2024). In addition, forest plantations play a crucial role in mitigating the effects of climate change. In southern and southeastern Mexico, fast-growing species such as white teak (*Gmelina arborea* Roxb.), teak (*Tectona grandis* L.f.), and eucalyptus (*Eucalyptus* spp.) have been widely used for timber production (Martínez-Zurimendi, 2015). In central Mexico, species from the *Pinus* genus are commonly planted, with *Pinus greggii* Engelm. ex Parl. being one of the most frequently used. This species is particularly valued for its ability to restore eroded soils, as well as its high potential for carbon sequestration, drought tolerance, adaptability to temperature fluctuations, and resistance to certain pests (López et al., 2017; Martínez-Sifuentes et al., 2020; Ortiz et al., 2021; Villegas-Jiménez et al., 2013). According to Carrillo-Castañeda et al. (2024), *P. greggii*, along with *P. pseudostrobus* and *P. devoniana*, demonstrated a high capacity for carbon sequestration when established for the restoration of severely degraded soils in Michoacán.

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Moreover, the increase in atmospheric CO_2 has triggered extreme weather events—such as floods, intense hurricanes, and others—resulting in both human and economic losses worldwide. In response, the Kyoto Protocol of 1997 requires developed countries to reduce their greenhouse gas emissions. It also offers flexible mechanisms, such as the Clean Development Mechanism (CDM), which promotes carbon capture and CO_2 sequestration through forest biomass (IPCC, 2014; Gómez-Guerrero, et al., 2021).

In the municipality of Huehuetla, there have been some attempts to establish small-scale plantations on private land, primarily to improve shade for coffee plantations and to obtain firewood. Arteaga and Castelán (2008) established an agroforestry plantation combining white teak (*Gmelina arborea* Roxb.), teak (*Tectona grandis* L.f.), and pink cedar (*Acrocarpus fraxinifolius* Wight & Arn.) with coffee. Their findings indicated that pink cedar and white teak were the species that adapted best to the site conditions. Huehuetla's altitudinal range spans from 400 to 1 300 meters above sea level and includes three different climate types, providing an opportunity to test various species with different temperature tolerances. According to Medina-Pérez et al. (2023), the Sierra Otomí-Tepehua —which includes the municipalities of Huehuetla, San Bartolo Tutotepec, and Tenango de Doria—is highly vulnerable to extreme hydrometeorological events. These are largely driven by deforestation and the conversion of forest land to agricultural and livestock uses, with landslides caused by heavy rainfall identified as one of the main regional hazards.

In light of these considerations, the current approach to forest plantations should be strategic, aiming to mitigate the effects of climate change—particularly through CO_2 capture, which is part of the payment for environmental services scheme (FAO, 2022; UNDP, 2023). This represents a local action with global impact. Consequently, there is a clear need to establish forest plantations and conserve natural forests. In the case of Huehuetla, Hidalgo, *Pinus greggii* appears to be a promising species, as it is well-suited to the municipality's natural conditions due to its subtropical characteristics (Perry, 1991; Ramírez-Herrera et al., 2005; Martínez-Sifuentes et al., 2020). Furthermore, Ortiz et al. (2021) recommend using *P. greggii* provenances from northern and central Mexico, where the species is endemic. Its genetic plasticity allows it to thrive on eroded soils, making it highly valuable for watershed protection and the restoration of degraded areas. Additionally, it exhibits rapid growth in such environments.

This implies the use of methodologies that address the degree of species adaptation to environments different from their natural distribution, such as climatic matching. In this sense, species distribution is limited by physiological, ecological, and biogeographic factors, which make it difficult to accurately determine a species' distribution area, as it is a dynamic phenomenon (Maciel-Mata, 2015). Climatic matching aims to expand the adaptation range of species from a different place of origin than the one where they are intended to be established, taking into account climatic parameters, with the most relevant being extreme temperatures, absolute minimum temperature, and the seasonal distribution and annual volume of precipitation (Golfari, 1963). Gómez (2021) discusses the concept of ecological matching, which is more complex, as it considers lithological, physiographic, climatic, and soil characteristics that influence the degree of adaptation. Therefore, considering that *P. greggii* is distributed along the mountain range of the Sierra Madre Oriental, where the municipality of Huehuetla, Hidalgo is located, the use of climatic matching will allow for the extension of the local distribution range of this subtropical conifer species.

Thus, the objective of this study was to establish and monitor an experimental plantation of *Pinus greggii* Engelm. ex Parl. in the municipality of Huehuetla, Hidalgo to evaluate initial growth, adaptation potential, and carbon capture.

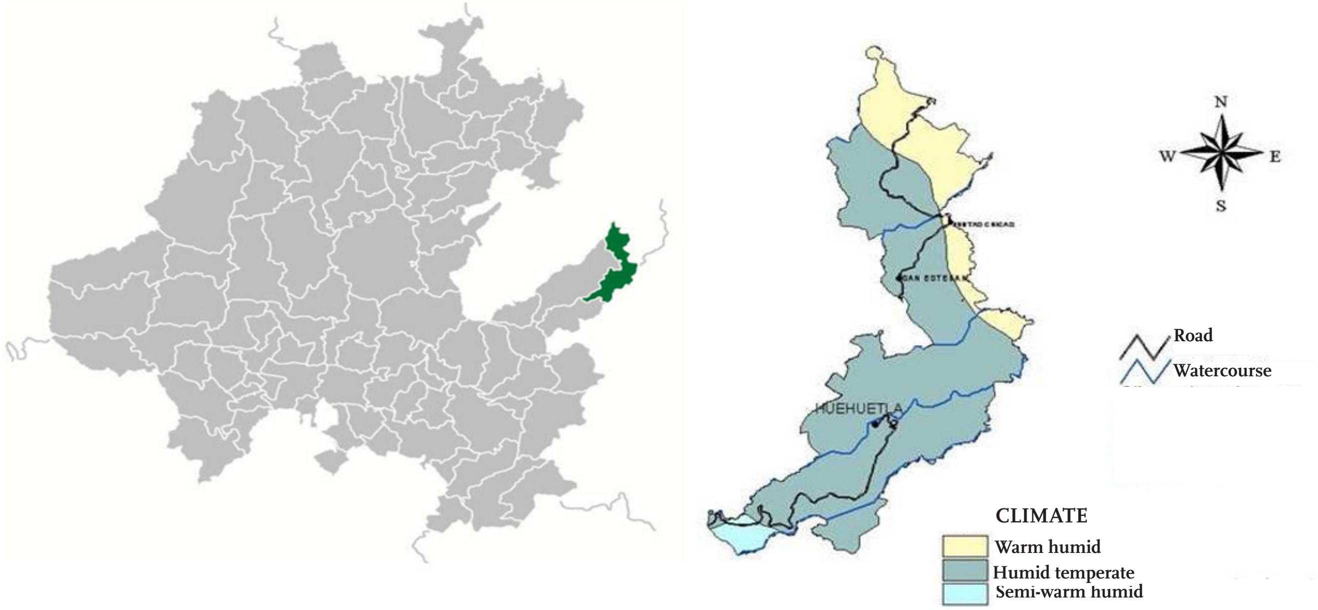
Materials and Methods

Study Area Description

The municipality of Huehuetla is located between 20° 23' and 20° 41' N and 97° 59' to 98° 10' W, situated in the northwestern and eastern part of the state of Hidalgo. It is part of the Tuxpan-Nautla region, within the coffee-growing area known as Otomí-Tepehua. According to García (2004), Huehuetla has three climate types: humid warm (Am) with summer rainfall, characterized by a short dry season during the cooler half of the year, yet with sufficient total annual precipitation to keep the soil moist year-round. This climate is intermediate between Af and Aw, resembling the former in total rainfall and the latter in annual distribution. The highest precipitation occurs in summer and part of autumn, when tropical cyclones affecting Mexico are most frequent, significantly increasing rainfall in this region. Other climate types present include humid sub-warm (ACf) with year-round rainfall, and humid temperate (Cm) with heavy rainfall during the summer. The average annual temperature ranges from 16 °C to 22 °C, with an average annual precipitation of 2 400 mm and elevation ranging from 400 to 1 300 meters above sea level. According to INEGI (2010), the predominant soil types are Umbrisol (56.95 %), Luvisol (36.2 %), Leptosol (4.32 %), and Phaeozem (2.33 %), with vegetation consisting of tropical rainforest and cloud forest. The plantation was established on Umbrisol soils, which, according to FAO (2025), are characterized by a dark layer rich in organic matter but with acidic pH (below 5.5) and low fertility. Figure 1 shows the location map of the municipality and the distribution of climate types.

Plantation Establishment

A total of 200 pine seedlings were planted, with an average height of 85 cm and a basal diameter of 8 mm, after



Source: Government of the State of Hidalgo (2010). Compilation based on INEGI data.

Figure 1. Location of the municipality of Huehuetla, Hidalgo, and climate.

having been grown in a nursery for two years. The site was prepared manually using the common pit system, since the slope of the terrain did not allow mechanization. Pits measuring 40 x 40 x 40 cm were dug to create favorable conditions for root establishment and to increase survival chances. The spacing between plants was 2.5 x 2.5 m in a square grid design. The plantation was established in June 2022 under a humid sub-warm climate. The seedlings originated from Estado de México.

Climatic matching

As a tool to assess adaptability to specific regions with particular climatic conditions, climatic matching was carried out using the FAO (1981) methodology, which considers the climatic variables of a species' place of origin in relation to the site where it will be introduced. Initially, the thermal profile of *Pinus greggii* was obtained. Then, the temperature data for the municipality of Huehuetla, reported by the National Meteorological Service, were evaluated and adjusted using the Regional Thermal Gradient for the planting site. The following climatic parameters were considered: minimum, optimal, and maximum temperatures, precipitation range, and altitude—for both the species and the plantation site.

The climatic requirements used for *P. greggii* included an altitudinal range of 1 400 to 2 600 meters above sea level, vital cardinal temperatures from 12 to 20 °C, with an optimal range of 16.8 to 17.5 °C, and extreme or lethal cardinal temperatures of 45 °C and -9 °C. Precipitation range between 640 and 1 370 mm, although it may extend from 500 to 2 900 mm (Dvorak et al., 1996; Ramírez-Herrera et al., 2005; Muñoz et al., 2012; Rodríguez et al., 2013). However, Martínez-Sifuentes et al. (2020) reported an average annual temperature of 25.4 °C in the distribution area in the Sierra Madre Oriental.

For the case of the municipality of Huehuetla, the thermal range considered was 14.0 to 32.5 °C, with an annual precipitation of 2 400 mm. The specific elevation of the planting site was 1 100 meters above sea level.

Evaluation and Monitoring

A total of four evaluations of survival and growth were conducted following the planting date, one every six months (December 2022, June 2023, December 2023, and June 2024). Tree measurement characteristics considered were height, basal diameter, and diameter at breast height (DBH). In the fourth evaluation, some trees reached or exceeded a height of 1.30 meters; for these, volume was calculated using the following formula:

$$V = \frac{\pi}{4} * d^2 * A * Cm$$

Where:

V = total tree volumen in cm^3

d = diameter at breast height (cm)

A = total tree height (cm)

Cm= form factor or morphic coefficient. This coefficient is used to correct the overestimation of tree volume that results from calculating it as a perfect cylinder. A value of 0.65 was used for conifers.

Initial Estimation of Carbon Content in Biomass

The amount of biomass in tree stands, expressed in terms of carbon content, is estimated to be approximately 50 %, regardless of ecosystem type, species, or region (Díaz et al., 2007; Acosta et al., 2009; Ronquillo-Gorgúa, et al., 2022). Similarly, the IPCC (2005) states that the carbon content in a tree is equivalent to 50 % of its biomass. Therefore, the carbon content for young trees is estimated using the following equation:

Biomass x Carbon content = Carbon stock.

Several methods are used to estimate carbon stock; however, all are subject to a certain degree of approximation. According to Portillo et al. (2023), the margin of error in biomass estimation—and therefore in carbon stock—ranges from 53 % to 80 %, regardless of the method used. Consequently, this study employed three estimation methods, including:

1) the method recommended by CONAFOR (Comisión Nacional Forestal), expressed by the following formula:

$C = V * FD * CC * BEF$

Where:

C: amount of carbon stored per tree

V: volume in m^3

FD: wood density factor; for conifers, the value is 0.48

CC: carbon content by species; for *P. greggii*, it is 51 %

BEF: biomass expansion factor; the value is 1.3

2) The Acosta et al. (2009) method, expressed with the allometric equation for the determination of biomass obtained for *Pinus patula* Engelm. Ex Parl. and other pine species given by:

$B = 0.0948 * DN^{2.4079}$

Where:

B: biomass (kg)

DBH: diameter at breast height (cm)

Multiplying the biomass by 0.50, which means that 50 % of the tree biomass is *C*.

3) The method proposed by the IPCC (2005) recommends multiplying the estimated tree volume by the carbon fraction, which is equivalent to 0.5.

Results and Discussion

The thermal table for *P. greggii* is presented in Table 1. This table made it possible to determine the species' adaptation percentage to the environmental conditions of Huehuetla, specifically at the plantation site, after subtracting the temperature differential (3.4 °C) resulting from the Regional Thermal Gradient.

An adaptation rate of 50 % to the local conditions was observed, which may be attributed to altitude as a natural limiting factor, given the 300-meter difference between the plantation site and the lower altitudinal limit of the species' natural distribution. However, as noted by Gómez (2021), although altitude is part of the physiographic characterization, it may also be influenced by factors such as lithology and soil properties. Similarly, Romahn-Hernández et al. (2020), in a study on the natural distribution of sacred fir (*Abies religiosa* (Kunth) Schltdl. et Cham.), reported high mortality rates among mature trees at the lowest altitudinal range. They also observed lower vigor in both mature and young trees, concluding that altitude is a key factor in the survival of sacred fir and suggesting an upward altitudinal shift in response to climate change.

In terms of temperature, the evaluated area falls within the thermal thresholds reported by Ramírez-Herrera et al. (2005), whose optimal range is between 16 and 18 °C. Regarding precipitation, the annual 2 300 mm recorded in Huehuetla falls within two of the reported ranges for *P. greggii*: 500 to 2 900 mm and 750 to 2 300 mm. Based on these favorable conditions, planting was carried out on June 26 and 27, 2022, when the soil was sufficiently moist following the onset of the rainy season. Initial measurements of the specimens—specifically, height and basal diameter—were taken on June 28, 2022, after having spent two years in the nursery.

The growth performance of the individuals is considered acceptable. After 24 months of monitoring and evaluation, 85 % of the 104 surviving trees exceeded 1.30 m in height. The average height was 1.85 m, with a maximum of 3.45 m (Figure 2). The average diameter at breast height was 1.14 cm, which allowed for an initial volume estimate of 147.5 cm^3. Additionally, the mean basal diameter exceeded 20 mm, with the largest reaching over 50 mm (Figure 3). Table 2 presents the estimated statistics, showing the percentage increase in both height and basal diameter two years after planting, relative to initial measurements.

Regarding survival, a gradual mortality of individuals was observed, with evaluation results showing survival rates of 85.5%, 79.5%, 67.5%, and 51% in the final assessment. Among the main factors contributing to mortality was pest damage, primarily by the stem borer (*Oncideres* spp.), which caused girdling of the stem and restricted sap flow in affected individuals. This was observed only during the first two evaluations. Additionally, mortality was attributed

Table 1. Thermal table generated for *P. greggii* Engelm. ex Parl., based on its cardinal temperatures.

Temperature range (°C)			Value
	Above 25.2		0
23.5		25.1	1
21.8		23.4	2
20.1		21.7	3
18.4		20.0	4
16.7	17.5	18.3	5
15.0		16.6	4
13.3		14.9	3
11.6		13.2	2
9.9		11.5	1
	Below 9.8		0

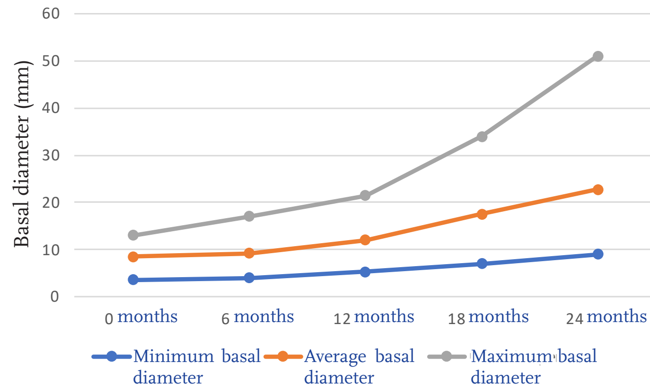


Figure 3. Basal diameter growth of *P. greggii* Engelm. ex Parl. in Huehuetla, Hidalgo.

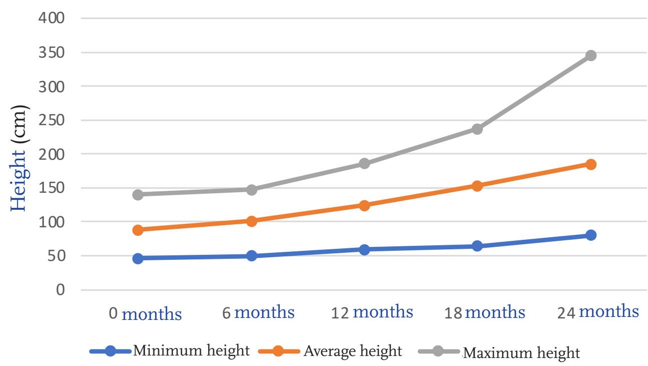


Figure 2. Height growth of *P. greggii* Engelm. ex Parl. in Huehuetla, Hidalgo.

to the exceptional and extreme drought that occurred in 2024, particularly during March, April, and May. This drought was driven by anticyclonic systems that caused heatwaves, resulting in hot to very hot conditions and a lack of rainfall. The drought affected 32 municipalities in the state of Hidalgo and nearly the entire national territory (CONAGUA-SMN, 2024). Despite these challenges, the survival percentage remained higher than the national average for reforestation efforts (36 %), although it fell short of the 80 % threshold required by the Comisión Nacional Forestal during the first year (Vázquez-Cisneros et al., 2018).

Ortiz et al. (2021) tested 13 provenances from northern and central Mexico on eroded soils in the Mixteca region of Oaxaca, finding high adaptability. Based on these results, the provenance from Estado de México was selected. Similarly, Flores et al. (2014) recommend the use of any provenance due to their strong performance, particularly as *P. greggii* is native to the Sierra Madre Oriental, including the state of Hidalgo (Domínguez-Calleros et al., 2017). Hernández-Martínez et al. (2007) also note that *P. greggii* Engelm. var. *australis* grows naturally in the municipality of Tenango de Doria, Hidalgo, which borders Huehuetla, thus potentially extending the species' adaptation range.

P. greggii has been used for its rapid growth and potential in programs aimed at the restoration of degraded soils and carbon sequestration. López et al. (2017) estimated the amount of carbon stored in soils associated with *P. greggii* and *P. oaxacana*, finding high levels of carbon both in the soil and leaflitter, and recommended their use on soils under extreme erosion conditions. Similarly, Muñoz et al. (2012) evaluated the survival and growth of *P. greggii* during the initial growth stage, finding that after six years, individuals exceeded five meters in height with a survival rate of 60 %.

In this context, the initial merchantable volume of pines was 1.475 x $10^4 \cdot m^3$ obtained after 24 months of monitoring. When expressed as carbon, this corresponds to 46.94 g per tree using the CONAFOR method. Using

Table 2. Estimated statistics for height and basal diameter variables.

Parameter	Variables					
	Initial height (cm)	Initial basal diameter (mm)	Height at 24 months (cm)	Basal diameter at 24 months (mm)	Height increase (%)	Basal diameter increase (%)
Minimum value	46	3.6	80	9	53.96	85.71
Mean value	90.1	6.9	184.7	22.7	68.85	106.75
Maximum value	140	11.3	345	51	84.53	127.45
Range	94	7.7	265	42	95.26	138.03
Standard deviation	19.689	1.612	52.134	8.046		

the method by Acosta et al. (2009), the estimated carbon content was 64.98 g, and according to the IPCC (2005) method, 73.5 g were obtained. From these results, an average of 61.81 g of carbon stored in the aboveground biomass of each tree was calculated, representing 226.84 g of CO_2 removed from the atmosphere. This conversion considers the molecular weight ratio between CO_2 (44) and carbon (12) (Jiménez et al., 2013; Robledo, 2015; Jáuregui et al., 2022). Furthermore, Pacheco et al. (2007) highlighted the importance of *P. greggii* for carbon capture, reporting a storage potential of 17.9 $t \cdot ha^{-1}$ during the first six years, equivalent to 65 $t \cdot ha^{-1}$ of accumulated carbon dioxide. These findings support the classification of this species as having high carbon sequestration potential. The variations in carbon storage estimates are attributed to the use of indirect methods, which calculate tree biomass using allometric equations or mathematical models based on regression analyses involving tree variables such as diameter at breast height (measured at 1.30 m), commercial (hc)and total height (ht), diameter growth, basal area, and wood specific gravity. Alternatively, estimates can be derived from stem volume, dry weight, and an expansion factor to calculate total tree weight. The "average tree" technique is also used, which assumes that the average-sized tree contains an average quantity of biomass (Fonseca et al., 2013; Fonseca-González, 2017).

Moreover, tropical species have also been used due to their important environmental role, as tropical forests can capture more than 40 $t \cdot ha^{-1}$ of CO_2 per year, equivalent to 2.3 to 10 t of carbon (Robledo, 2015). Aguirre et al. (2018) estimated that in an Andean forest in Ecuador, one hectare can capture 42.29 $t \cdot ha^{-1}$ of carbon accumulated in the tree, shrub, herbaceous, and necromass strata; when considering only the tree stratum, the value was 26.56 $t \cdot ha^{-1}$. Similarly, Seppänen (2002) reported that in southeastern Mexico, high-yield eucalyptus plantations (40 $m^3 \cdot ha \cdot yr$) can sequester between 320 and 610 $t \cdot ha^{-1}$ of CO_2 over a seven-year period, equivalent to 91 to 175 $t \cdot ha^{-1}$ of carbon. As early as 2007, the Ibero-American Network on Environmental Physics and Chemistry published several works on carbon capture in terrestrial ecosystems across Ibero-America, highlighting the growing need to evaluate this environmental service in economic terms (Gallardo, 2007).

The underlying principle is that forest ecosystems can capture significant amounts of greenhouse gases (GHGs), especially CO_2. For this reason, there has been increasing interest in recent decades in enhancing carbon stocks in vegetation through forest conservation, reforestation, and related strategies. Many studies have demonstrated the capacity of forest species to store carbon in their biomass (Rodríguez et al., 2016; Ronquillo-Gorgúa et al., 2022). This capacity has led to the development of payment schemes for environmental services related to carbon capture, contributing to forest conservation, soil recovery in deforested areas, and the restoration of degraded ecosystems.

Conclusions

After 24 months of plantation establishment, the increase in height has been greater than 90 cm, while basal diameter has increased more than 1.5 cm, and diameter at breast height exceeded 1 cm. After 12 months, 28% of the trees had surpassed the minimum height of 1.30 m required to measure diameter at breast height (DBH), reaching 85% by the final measurement. This growth has resulted in an initial volume that, when quantified as carbon stored per tree, exceeds 60 g.

The contribution of this species to mitigating climate change may be significant, as reforestation and afforestation efforts are a priority strategy for capturing excess atmospheric carbon dioxide while simultaneously promoting the conservation of native forests.

Local climate matchin is expected to expand the distribution range of *P. greggii*, since regional temperatures fall within the species' thermal tolerance. This makes it a promising candidate for reforesting unproductive or degraded lands requiring conversion to forest use.

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