



RESEARCH ARTICLE

OPEN ACCESS

Susceptibility of *Populus x euramericana* 'I-214' of Spanish origin to xylophagous attacks: durability tests for its possible inclusion in European standard

Eleana Spavento^{1*}, Mónica Murace¹, Luis Acuña², Silvia-Estela Monteoliva³ and M-Teresa Troya⁴

¹Wood Research Laboratory (LIMAD, as per initials in Spanish). School of Agrarian and Forestry Sciences (FCyF), National University of La Plata (UNLP), Buenos Aires, Argentina. ²Wood Technology Laboratory, Department of Agricultural and Forestry Engineering, University of Valladolid (UVA), Palencia, Spain. ³Institute of Plant Physiology (INFIVE), FCyF, UNLP-National Scientific and Technical Research Council (CONICET).

⁴Woods Protection Laboratory, Department of Forestry Products. Forestry Research Center (CIFOR). National Agrarian and Food Technology Research Institute (INIA), Madrid, Spain.

Abstract

Aim of study: to assess the natural durability of *Populus x euramericana* 'I-214' against xylophagous fungi and termites, and to carry out a macro-microscopic analysis of the alterations caused by each xylophagous agent in order to get the necessary information for its possible inclusion in existing European standards.

Area of study: a 20-years-old commercial plantation *Populus x euramericana* 'I-214' located in Quintanilla de Sollamas (42° 36' 00"N - 05° 49' 00" W), Spanish community of Castile-Leon.

Material and methods: material sampling and selection was carried out following EN 350:2017 for commercial sawn timber. Poplar resistance to xylophagous basidiomycete, soft rot fungi and subterranean termites was determined according to CEN/TS 15083-1:2005, CEN/TS 15083-2:2005 and EN 117:2012, respectively. The durability and use classes were estimated according to EN 350:2016 and EN 335:2013, respectively. The anatomical studies were carried out with Optical and Scanning Electron Microscope. Material characterization was carried out by reference to Anagnost (1998) and Schwarze (2007).

Main results: 'I-214' poplar wood proved to be "Not-durable" to the action of basidiomycetes, soft rot fungi and termites, use classes 1-2, and showed macro-microscopic evidence of these types of decay.

Research highlights: the information obtained in this study would allow the inclusion of clone I-214 in the standard EN 350 and its explicit classification within it.

Additional keywords: wood-decay fungi; termites; EN 350.

Abbreviations used: TM: test material; RM: reference material; RH: relative humidity; ML: mass loss; m_i: initial dry mass; m_f: final dry mass; DC: durability class; OM: Optical Microscope; SEM: Scanning Electron Microscope; TS: transverse section samples; LS: longitudinal section samples; CI: robust confidence intervals; F: fibre; V: vessel; h: hole; t: erosion trough; R: radial parenchyma cell; ep: erosion pitting; Fc: fungal colonization; fr: fracture; c: cavity; b: bore hole; m: mycelium.

Authors' contributions: conception, design and performance the experiments: ES and MTT. Wrote the paper: ES and MM. Conception and design of the research: ES, MM and MTT. All authors (ES, MM, LA, SEM and MTT) contributed reagents/materials/analysis tools/specific tests, participated in the analysis and interpretation of data/test, in the revision of the intellectual content and the drafting of the paper as well as read and approved the final manuscript.

Citation: Spavento, E., Murace, M., Acuña, L., Monteoliva, S.-E., Troya, M-T. (2019). Susceptibility of *Populus x euramericana* 'I-214' of Spanish origin to xylophagous attacks: durability tests for its possible inclusion in European standard. Forest Systems, Volume 28, Issue 2, e008. <https://doi.org/10.5424/fs/2019282-14660>

Received: 06 Feb 2019. **Accepted:** 05 Aug 2019.

Copyright © 2019 INIA. This is an open access article distributed under the terms of the Creative Commons Attribution 4.0 International (CC-by 4.0) License.

Funding: this work was funded by the EUROTANGO scholarship, in the frame of Erasmus Mundus Action 2-STRAND 1 - PhD Programme in the Universidad de Valladolid (2011-2014).

Competing interests: the authors have declared that no competing interests exist.

Correspondence should be addressed to Eleana Spavento: eleana.spavento@agro.unlp.edu.ar

Introduction

The planting of fast-growing species such as some *Populus* (poplar) species or clones has been carried out in several countries with the aim of obtaining raw material of economic, social and environmental interest

(Castro & Fragnelli, 2006; Kollert & Borodowski, 2014; Del Lungo, 2017). Within this context, Spain ranks among the 10 countries with the largest cultivated land areas and highest wood extraction of *Populus* spp. (Kollert & Borodowski, 2014). According to data from the Land Use and Crop Yield Survey (ESYRCE, 2018),

the total area of poplar plantations in this country is of approximately 122,502 ha, around 65% of which are located in the Castile-Leon region. The clone 'I-214', one of the most widely spread specie in the world (Garnica, 2017), represents over 50% of the total amount.

Broadly speaking, the use of poplar has traditionally been associated with the production of sheets and, with them, the manufacture of products such as matches, sticks, packagings and, mostly, plywood boards. Within the shredding industry, the use of this wood has been associated mainly with the manufacture of chipboards. Sawn products have traditionally been used for low added value timber products such as packing cases, crates and other products with little remanufacturing (Borodowski, 2017; Castro & Paganini, 2009; Baonza Merino & Gutiérrez Oliva, 2002).

However, the current market demands for more and more products with good physicochemical functionalities and well-known durability to meet the structural requirements of Eurocode 5. Wood and its derivatives are seen as good candidates, considering their environmental performance in terms of CO₂ sequestration (EN 1995-1-1:2016; Hildebrandt *et al.*, 2017).

Within this context, several researches have aimed at finding strategies to improve the structural capability of poplar wood, either solid wood or glued, in construction (Castro & Fragnelli, 2006; Casado *et al.*, 2012; Guillaumet *et al.*, 2014a, 2014b; IRAM 9662-4:2015; Rahayu *et al.*, 2015). In general, the use of wood entails exposure to situations conducive to decay, which compromise its service life (Ramage *et al.*, 2017). Consequently, knowing its natural durability - that is, its intrinsic resistance to biological decay caused by fungi, termites, beetles and marine borers (EN 350:2016) - allows for the definition of its ideal situation of use or protection degree according to use class, as specified in Eurocode 5 Design of Timber Structures (EN 1995-1-1:2016).

The standard EN 350:2016 sets out a list of the most prominent broadleaf species in the European construction sector and their corresponding natural durability class against biological agents. In this standard, the genus *Populus* is represented by the species *P. canescens*, *P. nigra*, *P. alba* and, without any specific data for the *P. hybrid* group, within which *P. x euramericana* 'I-214' could be considered to be included. Given the great number of I-214 poplars specimens existing in Spain and considering that the natural durability of its wood has not been strictly verified, it is of utmost interest to study its response to

xylophagous attacks to arrive at an early diagnosis for each agent so as to give it an explicit identification for its possible inclusion in the standard. In this sense, for use classes 2 and 3 brown and white rot fungi and wood borers stand out as the most destructive agents while for the use class 4 are the soft rot fungi and termites (Wilcox, 1978; Zabel & Morrell, 1992; Winandy & Morrell, 1993; Schwarze *et al.*, 2000; Schmidt, 2006; Schwarse, 2007; Murace *et al.*, 2010; EN 335:2013; Murace *et al.*, 2014). The current work was conducted to assess the natural durability of *Populus x euramericana* 'I-214' against xylophagous fungi and termites and to carry out a macro-microscopic analysis of the alterations caused by each xylophagous agent in order to get the necessary information for its possible inclusion in existing European standards.

Methods and materials

Wood material

We worked with *Populus x euramericana* 'I-214' -referred to as test material (TM)- from a 20-year-old commercial plantation located in Quintanilla de Sollamas (42° 36' 00"N - 05° 49' 00" W), Spanish community of Castile-Leon. The TM was received as 50 mm x 150 mm x 3000 mm beams which were conditioned in laboratory to achieve equilibrium moisture content at 12 + 2% (determined by digital hygrometer, Testo 606-1).

Material sampling and selection was carried out following EN 350:2016 for commercial sawn timber. In accordance with the minimum recommended ratio by the standard, out of the total of 100 beams, 40 were selected from different positions in the lot and reprocessed to obtain samples free from defects which were then sized according to the corresponding technical specifications. Samples were extracted from the inner section (material closest to the heartwood -identified by growth rings orientation¹) and outer section (material farthest from the heartwood) of each of the beams. Parameter followed for sample sectioning given that the selected beams of this commercial timber showed no evidence of duraminization and heartwood (or could not be observed/identified).

For basidiomycete fungi (brown and white rot) and termites, we used 15 x 25 x 50mm samples (standards CEN/TS 15083-1:2005 and EN-117:2012, respectively), and for soft rot fungi, 5 x 10 x 100mm samples (CEN/TS 15083-2:2005). For each section

¹Parameter followed for sample sectioning given that the selected beams of this commercial timber showed no evidence of duraminization and heartwood (or could not be observed/identified).

under study and type of decay, 40 repeats were carried out (320 repeats for TM).

As reference material (RM) we used *Fagus sylvatica* L. (beech) and *Pinus sylvestris* L. (pine), with similar characteristics to the TM, for fungi and termites, respectively. A total of 30 repeats for each type of decay were carried out (120 repeats for RM).

Establishment of durability test procedures

Basidiomycete fungi

Poplar resistance to xylophagous basidiomycete fungi was determined according to CEN/TS 15083-1:2005. The fungal species used were: *Coriarius versicolor* (L.) Quel. [= *Trametes versicolor* (L.) Lloyd], strain CTB 863 A, responsible for white rot, and *Coniophora puteana* (Shumach.) P. Karst., strain BAM Ebw.15, responsible for brown rot. Each strain was axenically cultivated in malt agar medium (1.5% agar, 3% malt) in 400 ml culture vessels and incubated at $22 \pm 2^\circ\text{C}$ and $70 \pm 5\%$ relative humidity (RH). Once the culture medium was covered by fungal tissue, the TM and RM samples were introduced into the culture vessels (2 TM or 2 RM x vessels) after determining their initial dry mass (m_i) and having sterilized them. After 16 weeks under controlled temperature and humidity ($22 \pm 2^\circ\text{C}$ and $70 \pm 5\%$ RH) in the culture chamber the RM samples were withdrawn from the vessels, conditioned (superficial mycelium extraction), and oven-dried at $103 \pm 2^\circ\text{C}$ to determine final dry mass (m_f). Subsequently, degradation was determined by loss in mass as a percentage of the initial dry mass (ML %), as calculated from Equation 1. Once a ML higher than 20% (established test reliability) was detected on this material, the same procedure was carried out for the TM samples (identified by sections). The criteria for determining durability classes (DC), according to ML

(%), are presented in Table 1. Use class was estimated according to standard EN 335:2013.

$$\text{ML (\%)} = \frac{m_i \text{ (g)} - m_f \text{ (g)}}{m_i \text{ (g)}} \times 100 \quad (1)$$

Where ML: mass loss, in %; m_i : initial dry mass, in g; m_f : final dry mass, in g.

Soft rot fungi

In this case, CEN/TS 15083-2:2005 was followed and the microbiota of the substrate was used to inoculate the test medium. A mixture of natural soil - fertile farm soil (20:80 ratio) with water holding capacity (WHC) ranging from 25 to 60% was used as substrate. The substrate was placed in 25 ml containers, enough to cover approximately 120 mm of the depth. The samples were dried in the oven and weighed to obtain their initial dry mass (m_i) before placing them in each container (20 TM samples identified per section and 10 RM samples), separated 20 mm from each other, and buried for 20% of their length. The containers were weighed and placed in a chamber under controlled conditions at $27 \pm 2^\circ\text{C}$ and $70 \pm 5\%$ RH for a period of 16 weeks. The initial reference weight (indirect indicator of system dehydration throughout the experiment) was recorded. In order to maintain the right water-holding capacity, each container was weighed weekly to keep the right humidity of the substrate. After the incubation period, 10 RM specimens were extracted at random in order to assess the validity of the test via $\text{ML\%} > 20\%$ (Equation 1). Once the test was validated, the same procedure was repeated for the TM samples. Durability class was calculated as the quotient x (Equation 2; Table 1: EN 350:2016) and use class for this material was determined based on this value (EN 335:2013).

Table 1. Durability class (DC) against the attack of xylophagous fungi and termites (EN 350:2016)

| DC | Description | ML ⁽¹⁾ | | DC | Description | Attack rating ⁽³⁾ | |
|----|--------------------|-----------------------|----------------------|----|--------------------|--------------------------------------|--|
| | | Basidiomycetes | Soft rot | | | Termites | |
| 1 | Very durable | ML < 5 | $x < 0.10$ | D | Durable | $\geq 90\%$ "0 or 1" and max 10% "2" | |
| 2 | Durable | $< 5 \text{ ML} < 10$ | $0.10 < x \leq 0.20$ | M | Moderately durable | $< 50\%$ "3, 4" | |
| 3 | Moderately durable | $10 < \text{ML} < 15$ | $0.20 < x \leq 0.45$ | | | | |
| 4 | Slightly durable | $15 < \text{ML} < 30$ | $0.45 < x \leq 0.80$ | S | Not durable | $\geq 50\%$ "3, 4" | |
| 5 | Not durable | $30 < \text{ML}$ | $x > 0,80$ | | | | |

⁽¹⁾ML (%): median mass losses; ⁽²⁾quotient of durability assessment (equation 2); ⁽³⁾90% of the test samples rated 0 or 1 and a maximum of 10 % of the test samples rated 2 and 0 % "3 and 4".

$$x = \frac{\text{median value of mass loss for timber test specimens}}{\text{median value of mass loss for reference timber test specimens}} \quad (2)$$

Where x: quotient of durability assessment.

Termites

The durability of wood exposed to subterranean termites was assessed according to standard EN 117:2012 using the species *Reticulitermes grassei* (Clément) as biological material. We used colonies consisting of 250 workers (those in molting stage -with whiter abdomens-, injured or immobile remained behind) and several nymphs and soldiers in a ratio of 1 to 5%, with reference to the proportion found in the colony the workers were extracted from.

Each colony was placed in 400 ml glass containers with a moist sand substrate (in proportion: 1 part of water to 8-10 parts of sand) and 0.5 g of farmed timber (*Pinus* sp. wood shavings) -initial food source of the colonies- evenly scattered on the approximately 40-60 mm thick sand layer. Subsequently, against one of the sides of each container (half-way up), a glass cylinder of approximately 1 mm wall thickness, height of 20 mm and diameter of 20 mm was introduced so that part of it extended out of the surface of the substrate. The containers were incubated at 28°C and 80% RH in the culture chamber for a period of 2-4 days, until the colony was uniform and lively.

In each container, and on the glass cylinder, a wooden specimen (TM or RM) was placed. Containers were then incubated in the culture chamber for 8 weeks, periodically checking the conditions of humidity of the substrate.

Once this period elapsed, all the specimens were removed, and after cleaning, a final examination was carried out. The number of living termites was counted (workers, nymphs and soldiers) and the attack ratings were visually analyzed (EN 117:2012): (0) no attack; (1) attempted attack, (2) light attack, (3) medium attack, and (4) heavy attack. The Survival Rate -SR- (%) was estimated using the ratio of the initial number of workers to the number of survivors.

The validity of the test was confirmed in 10 RM (pine) specimens through visual evaluation (attack level 4) and surviving workers count (SR: 50%). Even if it was not prescribed by the standard, ML (%) of the specimens was also assessed (Equation 1). The criteria for determining the resistance to deterioration caused by termites, durability classes (DC), according to attack ratings, are presented in Table 1 (EN 350:2016). Use class was estimated according to standard EN 335:2013.

Anatomical observations

Anatomical studies were carried out on TM (inner section) exposed to all xylophagous agents. We selec-

ted the material that showed mass loss percentages representative of the mean value obtained per type of test. The anatomical studies were carried out with an Olympus (CX31) Optical Microscope (OM) with a Lumenera's Infinity1 digital camera and a FEI (Quanta 200) Scanning Electron Microscope (SEM). For OM tests, transverse and longitudinal tissue sections (TS and LS) of the samples were stained with safranin at 1% and mounted in water temporarily. Unstained sections were observed under polarized light to detect loss of birefringence (indirect indicator of cellulolysis; diagnostic feature of brown rot). For observations with SEM, tissue sections without pretreatment were mounted on metal stubs using double-sided graphite tape and finally gold plated. Material characterization was carried out by reference to Anagnost (1998) and Schwarze (2007).

Statistical analysis

All statistical analysis was carried out with Software R©, 3.4.4 version (R Development Core Team, 2018). A total of 320 data values corresponding to each group (timber species, section under study and decay agents) were analyzed. Firstly, for all groups, statistical assumptions of independence, normality (Shapiro-Wilks test) and homoscedasticity (Bartlett test) were assessed. When the assumptions of normality and/or homoscedasticity were not achieved, the comparative analysis (ANOVA) common in classical linear statistics could not be used, then robust methods were applied to compare groups (Welch test). Bootstrap method was used to find mean and median confidence intervals and homogenous groups.

Results

Xylophagous fungi and termites

The results of macroscopic and microscopic evaluation of wood durability against basidiomycetes, soft rot fungi xylophagous and termites are shown in Table 2 and in Figures 1 to 4.

According to the results (Table 2), both inner and outer sections of poplar wood proved to be "Not Durable" (in according to the robust median CI value $\geq 30\%$ ML) to the xylophagous fungi employed, with statistically different, robust mean ML (%), between basidiomycete strains for each section under study (in uppercase) but without any significative difference in robust mean ML (%) between sections for both basidiomycetes and soft rot agents (in lowercase). Structural alterations, both macro and microscopic, were compatible with a low

Table 2. Durability test: results against different xylophagous agents according to mass loss (ML%).

| Xylophagous agent | Timber | Section | Mean ML (%)±CI ⁽¹⁾ | S-W ⁽²⁾ | Median ML ± CI (DC) ⁽³⁾ |
|-------------------------------|--------------------------------|---------|-------------------------------|--------------------|------------------------------------|
| <i>Trametes versicolor</i> | <i>P. x euramericana</i> I-214 | Inner | 55.75 ± 4.59 a A | 0.777 | 55.67 ± 7.19 (5) |
| | | Outer | 49.90 ± 4.57 a A | 0.254 | 51.67 ± 6.18 (5) |
| | <i>Fagus sylvatica</i> | MR | 44.15 ± 5.03 a A | 0.414 | 45.77 ± 3.97 |
| <i>Coniophora puteana</i> | <i>P. x euramericana</i> I-214 | Inner | 28.62 ± 3.09 b B | 0.977 | 28.36 ± 4.92 (5) |
| | | Outer | 29.73 ± 2.28 b B | 0.379 | 29.51 ± 4.76 (5) |
| | <i>Fagus sylvatica</i> | MR | 20.30 ± 4.06 c B | 0.053 | 18.18 ± 8.23 |
| Soft rot fungi | <i>P. euramericana</i> I-214 | Inner | 26.52 ± 2.33 a | 0.508 | 0.86 (5) |
| | | Outer | 28.46 ± 2.08 a | 0.091 | 0.82 (5) |
| | <i>F. sylvatica</i> | MR | 30.69 ± 2.08 b | 0.229 | |
| <i>Reticulitermes grassei</i> | <i>P. x euramericana</i> I-214 | Inner | 24.09 ± 3.46 a | 73.25 ± 5.05 a | 4 (S) |
| | | Outer | 23.41 ± 2.88 a | 74.64 ± 6.55 a | |
| | <i>Pinus sylvestris</i> | MR | 11.01 ± 0.35 b | 83.00 ± 5.49 a | 4 (S) |

⁽¹⁾ML (%): mass loss; CI: bootstrapping robust confidence intervals; significant differences between sections for each fungal strain are indicated by different letters in lowercase; significant differences between strains for each section are indicated by different letters in uppercase; Robust test ($p < 0,05$); ⁽²⁾S-W: Shapiro-Wilks test: non significant differences to normality test ($p > 0,05$); ⁽³⁾(DC): durability class; x: quotient of durability assessment, equation 2.

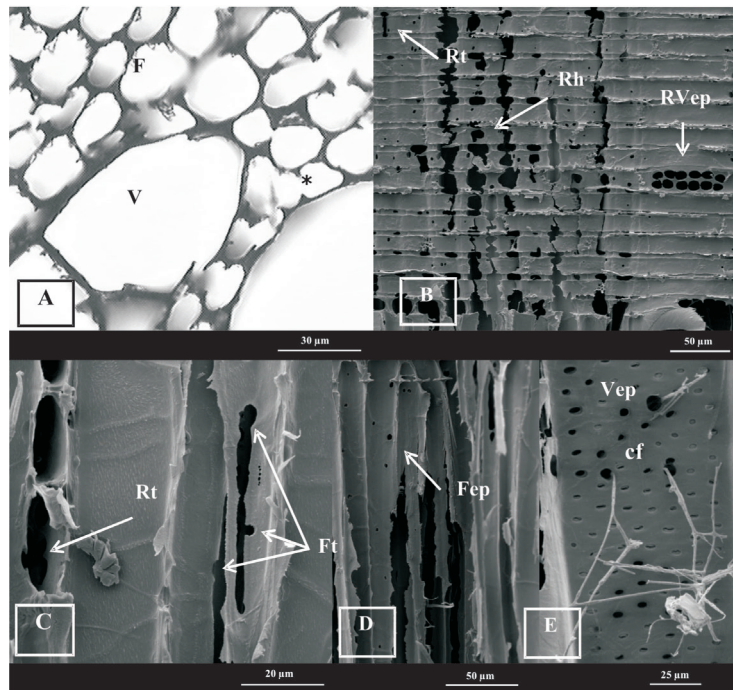


Figure 1. Diagnostic features of simultaneous white-rot. Photograph A: OM (TS): tissue deformation; wall thinning; thinning of fibre (F) and vessel (V); complete erosion (total thickness) of an area of the wall in fibre (*). Photographs B-E: SEM (LS). Photograph B: holes (Rh) and erosion troughs (Rt) radial parenchyma cell (R); erosion pitting at radius vessel level (RVep). Photograph C: erosion troughs in fiber (Ft); transversely oriented trough in radial cell (Rt). Photograph D: erosion pitting in fibre (Fep). Photograph E: erosion pitting in vessel (Vep); fungal colonization (cf) through pitting in vessels.

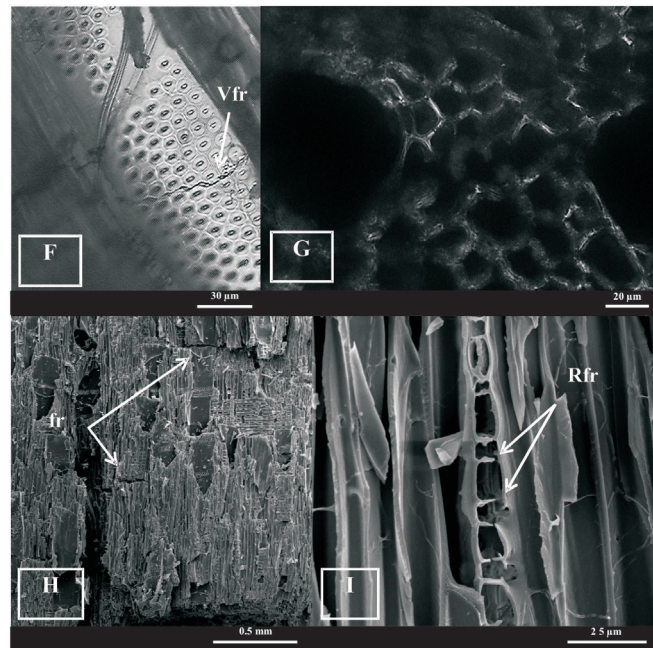


Figure 2. Evidence of brown rot. Photographs F-G: MO. A (LS): fracture in vessel (Vfr). Photograph G (TS): total loss of birefringence. Photographs H-I: SEM (LS). Photograph H: fractures (fr) perpendicular to the axial axis of the cells, overall. Photograph I: fractures in radial parenchyma cells (Rfr).

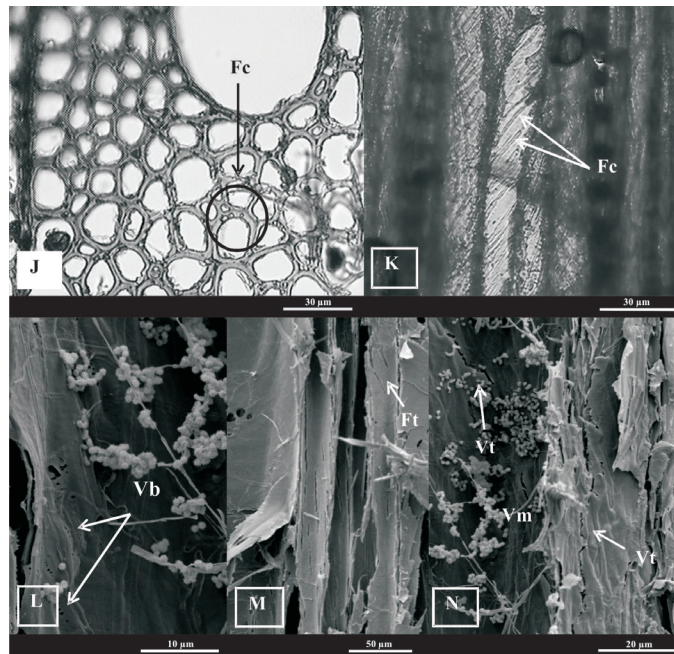


Figure 3. Diagnostic features of soft rot. Photographs J-K: MO. Photograph J (TS): round cavities in fiber walls (Fc). Photograph K (LS): cavities in a direction parallel to microfibrils in fibre (Fc). Photographs L-N: SEM (LS). Photograph L: bore holes (or small holes) in vessels (Vb). Photograph M: incipient erosion troughs in fibres (Ft). Photograph N: erosion troughs in vessels (Vt); mycelium in vessel (Vm).

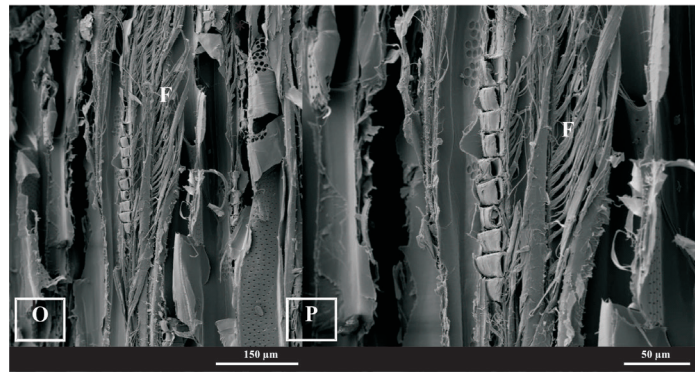


Figure 4. Microphotographs of evidence of damage by termites: Photographs O, P: SEM (LS): overall and detailed, respectively. Notice the tear in the wood, especially noticeable in fibers (F).

resistance to deterioration and were typical of each rot type (Schwarse, 2007; Anagnost, 1998).

Material exposed to white rot (*T. versicolor*) had characteristic fibrillar aspect and spongy consistency and was whitish in colour. OM observation revealed wall thinning in transverse tissue sections (TS) and consequent wall deformation in vessels, rays and fibers. In some areas, wall thinning affected the integrity of the middle lamella causing cell separation (Fig. 1A). Consequently, longitudinal section samples (LS) showed fibrillar aspect. SEM observation confirmed said fibrillar appearance and also revealed holes (Fig. 1B), erosion troughs (Figs. 1B y 1C) and pitting (Figs. 1B, 1D, 1E) in vessels, fibers and radial parenchyma. Vessels with abundant mycelium and evidence of fungal colonization in pitting (Fig. 1E) were also revealed.

Material exposed to brown rot (*C. puteana*) showed reddish-brown colouring, cubic fracture pattern and fragile consistency (it disintegrated into dust under pressure). OM observation showed strong contraction as well as fractures in vessels (Fig. 2F), rays and fibers. There was evidence of generalized loss of birefringence (Fig. 2G). Under SEM, the features described above -deformation and fractures- were observed (Figs. 2H y 2I), mycelium in vessels and fibers (less abundant than that observed in wood exposed to white rot fungi) and fungal colonization through the pitting in vessels.

Material exposed to soft rot showed spongy consistency and light brown colouring. OM observation of TS tissue revealed the presence of cavities in the fiber walls (Fig. 3J), which, in LS, were found in vessels as well. In both cases, parallel to the microfibril angle (Fig. 3K). Under SEM, and in LS samples, small holes (bore holes) were observed in vessels, most under 1µm (Fig. 3L). We also observed fibers and vessels colonized by mycelium with erosion troughs of serrated edges (Figs. 3M and 3N).

With reference to durability against attack by termites, results show that poplar is “Not durable” (EN 350:2016) in both sections under study (inner-outer), in accordance with what the standard EN 350:2016 describes for other species of the genus *Populus*. Survival rate and mass loss in both sections (inner and outer) confirmed results per attack level (only parameter for damage estimation considered by the standard).

Under SEM (Fig. 4), mechanical damage resulting from insect feeding was especially verified in fibers. This type of deterioration reduced the section of the specimens and, as a result, their quality and resistance to touch.

Discussion

Low durability of clone I-214 against several xylophagous agents coincides with what standard EN 350:2016 claims for several *Populus* species and also with what van Acker *et al.* (2003) claim for *Populus x euramericana*, even if they do not mention the clone specifically.

The similarity in its resistance to deterioration both in inner and outer sections is compatible with the age of the sampled specimens. Following Matyás & Pezslén (1994), around 30-40% of the wood under study could be considered juvenile and either lacked or had virtually nonexistent or non-distinguishable heartwood, according to what could be observed. The genus *Populus* in general, and the clone I-214 in particular, have white heartwood, which is non-distinguishable as a result of the absence of extractives, main indicator of durability. Hence, its heightened susceptibility to decay (Hillis, 1971; Bamber & Fukazawa, 1985; Taylor *et al.*, 2002; Martinuzzi, 2013). Even though we could not find any studies on poplar wood, several studies have described differential durability values between juvenile and

mature heartwood (Haupt *et al.*, 2003; Latorraca *et al.*, 2011) in other broadleaf species (*Tectona grandis* and *Robinia pseudoacacia*, respectively), suggesting that the source of such variance could be the presence of most extractives in the heartwood. Along these lines, in a study carried out to assess the role of extractives in naturally durable species, Kirker *et al.*, 2013 point out that extractive content is the main factor for durability.

As regards anatomical changes, samples showed the features to be considered in the diagnosis of white, brown and soft rot listed by Anagnost (1998) y Schwarze (2007). Extensive wall thinning, some cell separation, presence of bore holes and erosion troughs are typical features of simultaneous white rot, compatible with the spongy appearance and consistency found in the samples. This type of decay results from simultaneous degradation, in equal proportions, of the lignocellulosic matrix of the cell wall (Daniel, 2016). In brown rot samples, colouring, fragile consistency, contraction, presence of fractures and birefringence loss were consistent with preferred carbohydrate degradation. In this type of decay, lignin is only partially oxidized and is responsible for the brown colouring of the samples. Residual lignin and products resulting from carbohydrate polymerization prevent cellular integrity, and consequently tissue resistance diminishes (Schwarze *et al.*, 2000; Daniel, 2016). In both cases the presence of mycelium in vessels and pitting allowed for fungal colonization of wood (Schwarze *et al.*, 2000). In woods affected by soft rot, the presence of cavities is consistent with the removal of cell wall constituents near the hyphae, characteristic of this type of decay. However, lignin degradation was limited. Fungi responsible for soft rot degrade lignin partially just like those that cause brown rot, giving the wood its dark colour. In this type of decay, damage is usually superficial and appears on material exposed to extreme humidity conditions - wood in contact with the ground - but there is no significant loss of resistance like in the case of brown rot (Schwarze *et al.*, 2000; Daniel, 2016).

As regards damage caused by termites, Duarte *et al.* (2011) recorded mass loss of around 16% and attack level 4 in *Fagus sylvatica*; mass loss of 12% and attack level 4 in *Pinus pinaster*, and mass loss of 4.5% and attack level 3 in *Castanea sativa*, very dissimilar values (for the same attack level), and low when compared with normal values for tests of xylophagous fungi. One possible point of criticism to be made of the visual estimation recommended by the standard EN 117:2012 might be its subjectivity in the estimation of the attack level and corresponding use type. As a result of the low durability estimated and the alterations observed, the wood of this clone can only be used in very low

risk situations: indoor use, dry environment -or outdoor under cover, protected from severity of the weather-, without contact with the ground, use classes 1-2 (EN 335:2013), with a lifespan in extreme use situations - outdoor, in contact with ground - of under 5 years (IRAM 9600:1998).

Conclusions

Populus x euramericana 'I-214' 20-year-old wood of Spanish origin proved to be not durable to the action of the xylophagous organisms tested, which restricts its use to classes 1 and 2 (indoor use, dry environment -or outdoor under cover, protected from severity of the weather-, without contact with the ground). Even if its durability class coincides with that claimed for other clones and species of the genus, the values revealed in this test may be used as reference for its possible inclusion in the natural durability ratings for broadleaf species detailed in standard EN 350:2016, after completing the remaining tests for its characterization (*Anobium*, marine borers and *Thichoferus holosericeus*, if considered). Its inclusion within *Populus hybrid* would establish a specific identity for this clone, which would go in line with its worldwide distribution and commercial interest.

Acknowledgements

The authors wish to acknowledge technical staff and researchers from the National Research Institute and Agrarian and Food Technology (INIA, as per its initials in Spanish) and the Wood Technology Laboratory of the University of Valladolid (UVa), Spain, for the help provided during the research.

References

- Anagnost SE, 1998. Light microscopic diagnosis of wood decay. IAWA J 19(2): 141-167. <https://doi.org/10.1163/22941932-90001517>
- Bamber RK, Fukazawa K, 1985. Sapwood and heartwood: A review. Forestry Abstracts 46: 567-580.
- Baonza-Merino MV, Gutiérrez Oliva A, 2002. Rendimiento y calidades de chapa en clones de chopo a diferentes alturas del fuste. Investigación Agraria: Sistemas y Recursos Forestales 11(2): 311-323.
- Borodowski E, 2017. Situación actual del cultivo y uso de las Salicáceas en Argentina. Jornadas de Salicáceas-V Congreso Internacional de Salicáceas. 20 pp.
- Casado M, Acuña L, Basterra LA, Ramón-Cueto G, Vecilla D, 2012. Grading of structural timber of *Populus* ×

- euramericana clone I-214. *Holzforschung* 66(5): 633-638. <https://doi.org/10.1515/hf-2011-0153>
- Castro G, Fragnelli G, 2006. New technologies and alternative uses for poplar wood. *Boletín Informativo CIDEU* 2: 27-36.
- Castro G, Paganini G, 2009. New technologies and alternative uses for Poplar wood. *Boletín del CIDEU* 2: 27-36. ISSN 1885-5237.
- CEN/TS 15083-1, 2005. Durability of wood and wood-based products. Determination of the natural durability of solid wood against wood-destroying fungi, test methods-Part 1: Basidiomycetes. European Committee for Standardization, Bruxelles. 19 pp.
- CEN/TS 15083-2, 2005. Durability of wood and wood-based products. Determination of the natural durability of solid wood against wood-destroying fungi, test methods-Part 2: Soft-rotting micro-fungi. European Committee for Standardization, Bruxelles. 19 pp.
- Del Lungo A, 2017. Situación de Salicáceas en el mundo. *Jornadas de Salicáceas-V Congreso Internacional de Salicáceas*. 7 pp.
- Daniel G, 2016. Fungal Degradation of Wood Cell Walls. In: *Secondary Xylem Biology. Part II: Function and Pathogen Resistance of Secondary Xylem*, Chapter 8. 131-167 pp. Elsevier Inc Sweden. <https://doi.org/10.1016/B978-0-12-802185-9.00008-5>
- Duarte S, Ricart MG, Nunes L, 2011. Preferências alimentares de *Reticulitermes grassei* (Clément) (Isoptera: Rhinotermitidae) relativamente a diferentes espécies de madeira. *CIMAD. 1º Congresso Ibero-Latino Americano da Madeira na Construção*, Coimbra, Portugal. 7 pp.
- EN 1995-1-1, 2016. Eurocode 5: Design of timber structures - Part 1-1: General - Common rules and rules for buildings. European Committee for Standardization, Bruxelles. 150 pp.
- EN-117, 2012. Wood preservatives - Determination of toxic values against *Reticulitermes* species (European termites) (Laboratory method). European Committee for Standardization, Bruxelles. 22 pp.
- EN 350, 2016. Durability of wood and wood-based products - Testing and classification of the durability to biological agents of wood and wood-based materials. European Committee for Standardization, Bruxelles. 61 pp.
- EN 335, 2013. Durability of wood and wood-based products - Use classes: definitions, application to solid wood and wood based panels. European Committee for Standardization, Bruxelles. 14 pp.
- ESYRCE, 2018. Encuesta sobre Superficies y Rendimientos de Cultivos. Ministerio de Agricultura, Pesca y Alimentación de España. 178 pp.
- Garnica J, 2017. La importancia del chopo en la industria. *Jornadas de Salicáceas-V Congreso Internacional de Salicáceas*. 22 pp.
- Guillaumet A, Acuña Rello L, Piter JC, 2014a. Evaluación del rendimiento de las tablas de *Populus deltoides* 'Australiano 129/60' y 'Stoneville 67' para la fabricación de vigas laminadas encoladas. *IV Congreso Internacional de las Salicáceas en Argentina*. La Plata, Argentina. 8 pp.
- Guillaumet AA, Filippetti MC, Acuña Rello L, Piter JC, 2014b. Utilización de la madera de álamo en elementos estructurales. *XXXVI Jornadas Sudamericanas de Ingeniería Estructural*. Montevideo, Uruguay. 15 pp.
- Haupt M, Leithoff H, Meier D, Puls J, Richter HG, Faix O, 2003. Heart-wood extractives and natural durability of plantation-grown teak wood (*Tectona grandis* L.) a case study. *European J Wood Wood Products* 61(6):473-474. <https://doi.org/10.1007/s00107-003-0428-z>
- Hildebrandt J, Hagemann D, Thrän D, 2017. The contribution of wood-based construction materials for leveraging a low carbon building sector in Europe. *Sustainable Cities and Society* (Ed Elsevier) 34: 405-418. <https://doi.org/10.1016/j.scs.2017.06.013>
- Hillis WE, 1971. Distribution, Properties and Formation of Some Wood Extractives. *Wood Sci Technol* 5: 272-289. <https://doi.org/10.1007/BF00365060>
- IRAM 9662:4, 2015. Madera laminada encolada estructural. Clasificación visual de las tablas por resistencia. Instituto Argentino de Racionalización de Materiales. 16 pp.
- IRAM 9600, 1998. Preservación de maderas. Maderas preservadas mediante procesos con presión en autoclave. Instituto Argentino de Racionalización de Materiales. 23 pp.
- Kirker GT, Blodgett AB, Arango RA, Lebow PK, Clausem CA, 2013. The role of extractives in naturally durable wood species. *Internat Biodeterioration Biodegradation* (Ed Elsevier) 82:53-58 <https://doi.org/10.1016/j.ibiod.2013.03.007>
- Kollert W, Borodowski ED, 2014. Situación de las Salicáceas en el mundo. *Acta Jornadas de Salicáceas-V Congreso Internacional de Salicáceas*. 10 pp.
- Latorraca J, Dünisch O, Koch G, 2011. Chemical composition and natural durability of juvenile and mature heartwood of *Robinia pseudoacacia* L. *Anais da Academia Brasileira de Ciências* 83 (3): 1059-1068. ISSN online 1678-2690. <https://doi.org/10.1590/S0001-37652011005000016>
- Martinuzzi F, 2013. Fichas Técnicas de Madera. INTI Maderas y Muebles. *Revista CEMA* (114): 64.
- Matyás C, Pezslén I, 1994. Effect of age on selected wood quality traits of poplar clones. *Silvae Genetica* 46 (2-3): 64-72.
- Murace M, Spavento E, Keil GD, Saparrat M, 2010. Pudrición castaña: efectos sobre las propiedades de resistencia mecánica de la madera. *Quebracho-Revista de Ciencias Forestales* 18(1, 2): 37-46.
- Murace M, Spavento E, Rivas P, Saparrat M, Keil G, 2014. Comportamiento de *Pinus ponderosa* Dougl. ex. Laws. expuesto al hongo de pudrición castaña *Gloeophyllum*

- sepiarium (Wulf.: Fr.) P. Karst. *Quebracho Revista Ciencias Forestales* 22(1, 2): 114-124.
- R Development Core Team, 2018. R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <http://www.R-project.org>
- Rahayu I, Denaud L, Marchal R, Darmawan W, 2015. Ten new poplar cultivars provide laminated veneer lumber for structural application. *Annals of Forest Science* (Ed Springer). 14 pp. <https://doi.org/10.1007/s13595-014-0422-0>
- Ramage M, BurrIDGE H, Busse-Wicher M, Fereday G, Reynolds T, Shah D, Wu G, Yi L, Fleming P, Densley-Tingley D, *et al.*, 2017. The wood from the trees: The use of timber in construction. *Renewable and Sustainable Energy Reviews* (Ed Elsevier) 68: part 1: 333-359. <https://doi.org/10.1016/j.rser.2016.09.107>
- Schmidt O, 2006. Wood and tree decay. *Biology, Damage, Protection and Use*. Springer Verlag Berlin Heidelberg. 329 pp.
- Schwarse F, 2007. Review: Wood decay under the microscope. *Fungal biology reviews* 2(1): 133-170. <https://doi.org/10.1016/j.fbr.2007.09.001>
- Schwarze F, Engels J, Mattheck C, 2000. Fungal strategies of wood decay in trees. Springer Verlag. 184 pp. <https://doi.org/10.1007/978-3-642-57302-6>
- Taylor AM, Gartner BL, Morrell JJ, 2002. *Wood Fiber Sci* 34(4): 587-611.
- van Acker J, Stevens M, Carey J, Sierra-Alvares R, Militz H, Le Bayon I, Kleist G, Peek RD, 2003. Biological durability of wood in relation to end-use. *Holz Roh Werkstoff* (Ed Springer Verlag) 61: 35-45. <https://doi.org/10.1007/s00107-002-0351-8>
- Winandy J, Morrell J, 1993. Relationship between incipient decay, strength, and chemical composition of Douglas-Fir heartwood. *Wood Fiber Sci* 25(3): 278-288.
- Wilcox W, 1978. Review of literature on the effects of early stages of decay on wood strength. *Wood Fiber* 9(4): 252-257.
- Zabel RA, Morrell JJ, 1992. *Wood microbiology. Decay and its prevention*. Academic Press Inc 476 pp.