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Deformation induced by ethanol substitution in normal and tension wood of chestnut (*Castanea sativa* Mill.) and simarouba (*Simarouba amara* Aubl.)

Shan Shan Chang · Bruno Clair · Joseph Gril · Hiroyuki Yamamoto · Françoise Quignard

Abstract Wood deformation in the longitudinal and tangential directions induced by ethanol substitution and oven-drying was measured in normal wood (NW) and tension wood (TW) of chestnut (*Castanea sativa* Mill.) (TW with G-fibres) and simarouba (*Simarouba amara* Aubl.) (TW without G-fibres). The results show that with increased concentration of ethanol solution TW tends to contract, regardless of the presence or absence of G-fibres. In contrast, both NW and opposite wood expand at different rates in the longitudinal direction. These results are discussed and explained by the role of stress state at cell wall level.

Introduction

Usually the study of wood ultrastructure requires a preliminary sample preparation, such as resin embedding or CO_2 supercritical drying. These treatments require dehydration of the wood samples with an ethanol series, after which the observed structures are expected not to be modified. However, recent macroscopic

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deformation measurements after ethanol serial substitution show that tension wood (TW) with G-fibres behaves very differently to normal wood (NW) (Clair et al. 2008), thus further focus was placed on this particular question.

TW is a peculiar wood tissue that most frequently forms on the upper side of leaning trunks or branches in hardwood species (Isebrands and Bensend 1972). However, Onaka (1949) as well as Fisher and Stevenson (1981) also reported occasional TW formation on the lower side. TW usually occurs in the region of fastest growth in eccentric stems or branches (Wardrop 1964). It tends to contract during maturation and generates high tensile stress (Wardrop 1964; Fisher and Stevenson 1981) that assists stems or branches to maintain or recover a preferred orientation (Kuo and Timell 1969). In many hardwood species such as beech, poplar and chestnut (*Castanea sativa* Mill.), TW contains fibres with a special morphology and chemical composition due to the development of the so-called gelatinous layer (G-layer) (Onaka 1949) that replaces the S₃ layer and a part or the whole of the S₂ layer (Wardrop and Dadswell 1955). Although it has been well established that G-layer is the driving force of stress generation (Yamamoto et al. 2005; Fang et al. 2008), it is also well known that some species without G-layer, e.g. simarouba, still exhibit high growth stresses (Ruelle et al. 2007a, b).

The drying shrinkage due to the variation of wood moisture content below fibre saturation point has already largely been described in the literature, but in most cases, it concerned NW shrinkage due to dehydration (evaporation) (Skaar 1988). Some studies have been done on the wood deformation behaviour in organic solvent (Ashton 1973; Kajita et al. 1979; Mantanis et al. 1994), but they addressed the evaluation of various possible predictive factors for swelling of wood in organic liquids from a dry state to equilibrium state of moisture. Ishimaru et al. (2001a, b, c) performed a serial study on mechanical properties changes of wood swollen in organic liquids. The present study addresses the deformation induced by ethanol substitution with special interest on TW behaviour compared to NW and on the role of G-layer in its behaviour.

Materials and methods

Experiments were performed on a tilted stem of chestnut (*Castanea sativa* Mill.) and a branch of simarouba (*Simarouba amara* Aubl). Both were around 5 cm in diameter. Chestnut produces TW with a typical G-layer and exhibits high longitudinal shrinkage up to ten times higher than NW (Clair et al. 2003a); simarouba has been well described by Ruelle et al. (2007a, b). This species does not develop G-fibres even when growth stress measurements indicate the TW production. Longitudinal shrinkage in simarouba TW is known to be two times higher than in NW (Ruelle et al. 2007a, b).

Wood samples were extracted from both sides of the axes (Fig. 1a, b): upper side (marked as U; TW in chestnut is easily identified by its brown colour) and lower side (marked as L). The cross-section of simarouba branch shows distinct eccentricity (Fig. 1a).



Fig. 1 Cross-section of simarouba (a) and chestnut (b), showing the approximate positions of the samples and c sample preparation for both species (chestnut taken as example)

A batten (8–10 mm width) was taken through the pith and was sawn into six longitudinal strips successively along radial direction, 2 mm in radial (*R*) direction and 7 mm in tangential (*T*) direction, from bark to pith for both upper side and lower side (Fig. 1c). They were marked as CU1–CU6 for chestnut upper side and CL1–CL6 for lower side, respectively (the same method for simarouba, but only four strips for SL due to the narrow distance from pith to lower side). Each strip was then cut into six samples along the longitudinal (*L*) direction on a circular mini-saw (only the first and the fourth samples were used for this study). The precise dimension of the samples is $2 \times 7 \times 50 \text{ mm}^3$ ($R \times T \times L$) after polishing. Thus, 24 samples for chestnut and 20 samples for simarouba were obtained.

In this way, wood samples were divided into two paired sample sets: one set of samples was dehydrated by serially increasing the concentration of ethanol solution (25, 40, 55, 70, 85, 95 and 100%). The other set was dried by heating at 102°C for 72 h in a ventilated oven. The two sets were 10 cm apart from each other (samples in-between were dehydrated with other solvents and results will be presented in a separate article).

Deformation calculation

For each sample, the measurements were performed using a digital micrometer (precision 0.001 mm). Dimensional changes of samples due to ethanol substitution in *L* and *T* directions are represented as ε_L and ε_T

$$\varepsilon_L(\%) = (L - L_0) \times 100/L_0 \quad \varepsilon_T(\%) = (T - T_0) \times 100/T_0,$$

where L_0 and T_0 are the longitudinal and tangential dimensions in green condition, and L and T are the dimensions in serially dehydrated condition. According to the formula, a positive value of ε_L or ε_T indicates that the sample expands, and a negative one indicates shrinkage.

The deformation of samples due to oven-drying (α_L, α_T) was quantified by a shrinkage ratio, now counted positively in the case of shrinkage

$$\alpha_L(\%) = (L_W - L_D) \times 100/L_W \quad \alpha_T(\%) = (T_W - T_D) \times 100/T_W,$$

where L_W and T_W are the longitudinal and tangential dimensions in green condition, and L_D and T_D are the dimensions in oven-dried condition.

Microfibril angle (MFA) measurement

After ethanol serial dehydration and oven-dry measurements, mean microfibril angle (MFA) was measured by X-ray diffraction method using small test specimens of $1 \times 1 \times 15 \text{ mm}^3$ ($R \times T \times L$). MFA was calculated using the improved Cave's method (Cave 1966; Yamamoto et al. 1993) for chestnut and the specific correction factor proposed by Ruelle et al. (2007a) for simarouba.

Results and discussion

Table 1 summarises all measurements done on simarouba and chestnut samples: L and T deformations induced by serial ethanol substitution, oven-dry shrinkage and microfibril angle (MFA) of the ethanol substitution and oven-dry samples.

Identification of wood types

The presence of TW can be quantitatively estimated at the periphery of the stem by growth strain measurements (Fournier et al. 1994; Yoshida and Okuyama 2002). On the other hand, because of the (sometimes unpredictable) history of the stem, it is difficult to know whether the core of the log is TW or NW. Staining (Grzeskowiak et al. 1996) has been used to distinguish TW from NW, but it was shown that one of the best predictors is the mapping of longitudinal shrinkage (Clair et al. 2003b; Gril et al. 2003, Ruelle et al. 2007b). Therefore, the duplicate sample was used to confirm the presence and severity of TW by measuring longitudinal shrinkage. The measurement of MFA on both of the paired samples allowed the conclusive definition of the wood type.

Table 1	ummary	of L and	T defoi	rmation	s after se	srial etha	nol subs	stitution,	oven-dry	/ shrinka	ge and	microfi	bril an	gle (N	IFA) fc	ır simar	ouba a	nd chestr	Iut	
Species	Sample	$\epsilon_L (\%_0) $	luring e	thanol se	eries				α_L (%)	ET (%0) (during et	thanol s	eries				α_T	MFA _{eth}	MFA _{od}	Wood
		25%	40%	55%	70%	85%	95%	100%		25%	40%	55%	70%	85%	95%	100%	(200)	C	C	iype
Simarouba	SUI	0.06	0.14	0.30	0.30	0.28	0.26	0.22	1.72	0.75	1.18	1.94	3.34	3.34	1.94	2.91	38.3	30.8	28.0	MM
	SU2	0.30	0.44	0.50	0.68	0.64	0.58	0.56	1.32	0.54	0.86	2.48	3.99	4.21	3.45	2.80	42.2	31.1	29.2	MN
	SU3	0.14	0.36	0.52	0.70	0.74	0.52	0.54	1.24	0.65	1.08	2.48	3.55	3.12	3.12	3.34	39.6	32.6	30.6	ΜN
	SU4	0.54	0.80	1.22	1.36	1.44	1.34	1.22	2.49	0.54	0.75	1.94	3.02	4.74	2.59	2.69	40.3	34.5	37.8	MO
	SU5	0.64	1.19	1.59	1.85	1.95	1.89	1.83	1.75	-0.11	0.00	1.51	2.58	2.26	0.11	2.04	39.1	38.1	37.1	MO
	SU6	0.30	0.84	1.40	1.68	1.64	1.58	1.30	2.37	0.32	1.08	2.48	3.56	3.23	3.34	2.91	38.7	40.4	37.0	MO
	SLI	-0.10	-0.16	-0.34	-0.28	-0.40	-0.66	-0.64	2.84	0.99	1.86	3.95	5.26	4.82	4.28	3.51	49.6	22.2	31.5	
	SL2	-0.08	-0.12	-0.32	-0.20	-0.30	-0.40	-0.66	3.82	0.11	0.87	2.51	3.81	3.81	0.65	3.16	47.8	28.1	22.2	ΜT
	SL3	-0.06	-0.18	-0.02	-0.04	-0.14	-0.18	-0.36	3.98	2.07	2.40	4.14	5.88	4.57	6.32	4.36	47.0	29.0	23.8	ΤW
	SL4	-0.16	0.08	0.10	0.26	-0.02	0.08	-0.26	3.56	0.87	1.74	2.72	4.03	2.83	3.05	2.72	44.0	24.2	22.8	ΤW
Chestnut	CU1	-0.30	-0.28	-0.16	-0.06	-0.22	-0.32	-0.36	6.14	0.60	1.33	3.02	3.99	3.75	3.26	3.50	73.2	13.7	14.3	ΜT
	CU2	-0.02	-0.32	-0.30	-0.30	-0.66	-0.62	-0.70	7.60	0.72	1.45	3.62	4.58	3.74	2.65	3.37	121.2	10.2	8.4	ΜT
	CU3	0.04	0.02	0.06	0.30	-0.12	-0.14	-0.06	5.99	0.60	1.20	4.69	4.57	3.13	1.44	3.49	94.2	7.6	7.4	ΜT
	CU4	-0.14	-0.22	-0.32	-0.54	-0.56	-0.62	-0.66	6.06	0.12	0.48	3.23	4.19	3.59	2.99	1.92	118.3	8.4	9.0	ΜT
	cu5	0.14	-0.16	-0.40	-0.22	-0.42	-0.36	-0.58	6.49	1.33	2.53	2.29	2.65	3.13	4.58	1.33	135.3	7.5	8.6	ΜT
	CU6	0.52	0.82	0.80	0.60	0.54	0.52	0.48	3.63	1.08	2.16	5.63	6.59	3.35	3.47	2.99	87.8	16.7	28.5	
	CLI	-0.10	0.10	0.26	0.32	0.36	0.42	0.20	2.54	-0.37	-0.24	1.46	3.78	1.34	0.00	1.59	60.6	21.0	30.5	
	CL2	-0.02	0.06	0.42	0.50	0.66	0.42	0.32	1.19	0.85	1.46	3.90	5.36	3.90	2.80	4.14	56.4	24.1	26.8	ΜN
	CL3	-0.02	0.16	0.38	0.54	0.74	0.46	0.28	1.23	1.95	2.43	3.65	5.60	5.72	4.26	2.31	60.3	25.3	27.7	MN
	CL4	-0.08	0.26	0.44	0.82	0.70	0.90	0.60	-0.72	0.61	0.97	3.03	4.00	2.91	3.03	2.55	70.4	23.7	26.9	MN
	CL5	-0.04	0.32	0.66	06.0	0.92	0.84	0.64	-1.43	0.97	1.57	3.39	4.72	3.02	2.06	2.54	63.2	23.4	25.2	MN
	CL6	0.40	0.64	1.22	1.32	1.28	1.34	1.30	0.14	2.40	3.13	4.93	4.57	4.81	0.48	4.09	75.2	15.1	25.9	
Values rem	oved for 1	ater analy	sis are	in italics																
$\varepsilon_L (\varepsilon_T)$ defo NW normal	rmation cl wood	iange due	to etha	nol subst	itution; o	$x_L (\alpha_T) dr_L$	ying shri	nkage; M	IFA _{eth} (M	FA _{od}) MI	∃A of etl	hanol (c	or oven	-dry) sl	pecimer	ls; <i>TW</i> t	ension	wood; OV	/ opposite	;pood;



Fig. 2 Relation between microfibril angle of oven-dry samples (MFA_{od}) and longitudinal shrinkage (α_L) of simarouba (**a**) and chestnut (**b**). *Filled symbols* SU and CU; *empty symbols* SL and CL

The lower side wood of simarouba had higher longitudinal shrinkage than the upper side wood, while the reverse was true for chestnut (Table 1). This would indicate that TW is located on the upper side (as expected) in chestnut, whereas it is on the lower side in simarouba.

Figure 2 shows the relation between α_L and MFA of oven-dry samples in simarouba and chestnut. The upper side wood of chestnut exhibited low MFA but high α_L value, which agrees with the physical properties of TW. These results allowed final classification of the wood types: the upper side wood of chestnut was identified as TW and the lower side wood as NW. The properties of the sample CU6 (near pith) were close to the ones of the lower side wood.

Simarouba shows comparatively high α_L with a lower MFA (around 20°) on the lower side (SL2, SL3, SL4), and small α_L (around 0.1%) and larger MFA (around 25°) on the upper side (SU1, SU2, SU3). At positions SU4, SU5 and SU6, comparatively high α_L and very high MFA (35°) were measured. Longitudinal shrinkage of lower side wood of simarouba, even if not as high as in chestnut TW, is twice the one of upper side wood (SU1, SU2, SU3), which corresponds to what was observed for the TW of this species (Ruelle et al. 2007b).

Based on the good agreement of our measurements with observations done by Ruelle et al. (2007a, b) on simarouba TW and NW, samples SL2, SL3, SL4 were classified as TW, although the MFA value is higher than that expected in TW. The positions SU4, SU5, SU6 which are opposite to TW were classified as opposite wood (OW) and the positions SU1, SU2, SU3 as NW. Unusual repartition of TW has already been observed by Fisher and Stevenson (1981). Here, the repartition of wood types could be explained by an active process of the tree for the light search in the competitive context of tropical rain forest. Samples SL1 and CU6 could not be attributed to a clear wood type, so they have been removed for further analysis.

Because the samples used for ethanol substitution and oven-dry shrinkage were not located at exactly the same position, MFA of the two sets of samples were measured in order to control the identification of wood types (Fig. 3). This led to the removal of two other samples, CL1 and CL6, where MFA of oven-dry sample was



Fig. 3 Microfibril angle of the ethanol substitution (MFA_{eth}) and oven-dry (MFA_{od}) samples of simarouba (a) and chestnut (b)

not well paired with ethanol samples. Comparison of MFA also confirmed the need to remove SL1 and CU6.

Deformation induced during ethanol substitution

Along the *T* direction during serial ethanol substitution, no clear difference was observed between NW/OW and TW of both simarouba and chestnut (Table 1). Most of the values of ε_T were positive which meant that the specimens expanded in *T* direction. The largest ε_T value appeared around 70% ethanol solution. At higher concentration, the expansion was partly recovered.

Figure 4 shows the average ε_L of the different wood types in simarouba and chestnut. As expected with regard to the molecular size of ethanol compared to water, NW swelled during substitution and this swelling became even higher when MFA highly increased in OW.

Considering the same mechanism, in TW the orientation of MFA is low in $G(S_2)$ so the contribution to longitudinal dimension change should be essentially affected



Fig. 4 Mean longitudinal deformations (ε_L) of different wood types in simarouba and chestnut after serial ethanol substitution. *Error bars* show 95% confidence intervals

by changes in the other layers (OL: middle lamella primary walls and S_1 layers) which have high MFA. Consequently, TW should slightly swell, whereas shrinkage was observed.

Since this shrinkage occurred both in chestnut and simarouba TW, it cannot be attributed to the peculiar morphology and behaviour of the gelatinous layer. The explanation needs to be based on a feature shared by all types of TW, namely their tensile stress. The following interpretation is proposed.

The formation of TW is characterised by the development of considerable growth stresses (Wardrop 1964; Fisher and Stevenson 1981; Okuyama et al. 1994). Even after the release of macroscopic stress resulting from wood sample cutting, some residual stresses remain in the microstructure (Fig. 5). Presumably, the G (S₂) layer is in a state of tension relatively to other layers (S1 and CML) which, in order to ensure a static balance, needs to be subjected to large compressive stress (Archer 1987). The dehydration by ethanol could provoke a drastic increase of the molecular mobility within the amorphous substance of the cell wall, similar to that induced by moisture change under load (Back and Salmén 1982). Concerning L stresses, this softening effect should be less considerable in the G/S₂ layer, where the microfibrils are more axially oriented than in the S1/CML, where they are mainly oriented transverse to the fibre axis. As a result, the latter undergo a large inelastic shrinkage or compression set, while the balance of internal stresses within the cell wall is shifted in favour of the release of the tension in the G/S₂ layer, resulting in the observed macroscopic shrinkage. In TW without G layer, where the microfibrils of the S₂ layer are not oriented as axially as in a G layer, some inelastic extension is likely to occur in the S2 as well partly reducing the shift of balance and the resulting macroscopic strain.

This interpretation could also explain the paradoxical longitudinal shrinkage of the TW of many hardwood species without G-layer which is commonly two times higher than in NW despite the lower MFA (Ruelle et al. 2007b) (as it was here



Fig. 5 Schematic representation of the stresses change before and after ethanol serial substitution. $G(S_2)$ G-layer (or S_2 layer); *OL* other layers (including S_1 layers, primary walls and middle lamella); *r* and *t* are the directions respectively radial and tangential to the fibre

observed in simarouba). The loss of bound water induces a highly anisotropic shrinkage, very large transverse to the fibres but normally prevented in the L direction by the more or less axial orientation of the crystalline microfibrils. In TW, the unusually high level of residual stress would be responsible for the same kind of compression set of the other layers, as the one described above as a result of ethanol substitution. In TW with G-layer, longitudinal shrinkage can be even higher and can reach up to eight times the one of NW, because of the gel structure of its secondary wall (Clair et al. 2008).

Conclusion

Longitudinal deformations induced by ethanol substitution clearly discriminate wood types between TW, which tends to contract, whilst NW and OW tend to expand. The fact that either with or without G-layer, TW tends to contract shows that this behaviour is not linked to the peculiar structure of the G-layer, and an explanation based on the role of stress state at cell-wall level is proposed.

This study only throws some light on this interesting problem. Obviously, there is still a lot to do in order to better understand how this deformation behaviour is produced in ethanol solution. Furthermore, it is not known whether there exists the same deformation behaviour in other organic solvents or other species. This should be investigated in the future on more species and more specimens per species. The next step in this study will be concerned with the anatomical structure, chemical composition of cell wall components and material physical properties in order to fully understand the phenomena.

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