# On the detachment of gelatinous layer in tension wood fiber

Bruno Clair, Bernard Thibaut, Junji Sugiyama

**Bruno Clair . Bernard Thibaut . Junji Sugiyama**

# On the detachment of gelatinous layer in tension wood fibre

Accepted as original article in ***Journal of Wood Science*** (6th may 2004)
Will be probably published in volume **51**(2) 2005

**Abstract** The detachment of gelatinous layer (G-layer), often observed on microtome cross sections, leads some authors believe that G-layer cannot act as the driving force of longitudinal shrinkage in tension wood. The aim of this study was to observe the detachment of G-layer along fibres. Green wood block was cut transversely into two samples. One sample was kept in water and the other oven-dried. One face being common to both samples, detachment of G-layer has been studied on the same fibres. Observations have been performed after blocking deformation by embedding. It reveals that the detachment of G-layer is a cutting effect produced during the first making of the transverse face of the wood block to be embedded. After 100 µm far from this primary surface of the sample no detachment can be observed. Drying shrinkage does not affect or little this detachment. The result seems to explain well why the detachment of G-layer occurs during sectioning using a conventional sliding microtomy. These observations prove the adhesion of G-layer in massive wood and confirm the active role of G-layer in tension wood properties.

**Key-words** Wood cell wall, cutting effect, gelatinous layer (G-layer), growth stress, tension wood, *Populus euramericana*

B. Clair[1] (corresponding. author.). J. Sugiyama
Laboratory of Cell Structure and Function, Wood Research Institute, Kyoto University, Uji-Kyoto 611-0011, Japan

B. Thibaut
Laboratoire de Mécanique et Génie Civil (LMGC), UMR 5508 CNRS – Université Montpellier 2, Place E. Bataillon, CC 048, 34095 Montpellier CDX 5, France

*Present adress:*
[1]Laboratoire de Mécanique et Génie Civil (LMGC), UMR 5508 CNRS – Université Montpellier 2, Place E. Bataillon, CC 048, 34095 Montpellier CDX 5, France
Tel: +33-467-14-34-83 - Fax: +33-467-14-47-92
e-mail : clair@lmgc.univ-montp2.fr

## Introduction

To maintain verticality, most angiosperms are able to produce highly tensile stressed wood on the upper side of the leaning trunk. The stress asymmetry between both upper and lower sides of the trunk permits then to bend it to recover verticality[1, 2]. This xylem with high tension stress is called tension wood. It is characterised in many species by an unusual cell wall structure with a characteristic layer called gelatinous layer (G-layer)[3]. G-layer is known to have a high cellulose content with a high degree of crystallinity[4, 5] and cellulose microfibrils are oriented along the axis of the cell[6]. These differences in chemical composition and structure give to macroscopic tension wood some particular properties in comparison to normal wood, notably a high shrinkage[7-11]. These high macroscopic shrinkages can find explanations in the properties of G-layer itself. In spite of its structure with microfibrils axially oriented, G-layer is subject to high shrinkage both in transverse[5] and longitudinal directions[12]. However, in order to contribute to the macroscopic behaviour, G-layer has to have a relatively higher elastic modulus in its axial direction and to be in tight adherence with the other layers of the cell. G-layer being often observed loosely attached to normal secondary wall layer[13-15], its contribution to macroscopic behaviour, especially to axial shrinkage, has been put in question[5, 16]. The aims of this study were to observe, after blocking deformation by embedding, the detachment of the G-layer from $S_2$-layer along the fibre. Observations were made on never-dried wood and on dried wood in order to evaluate the influence of drying on the G-layer detachment.

## Materiel and methods

Plant material

Experiments have been performed on poplar tension wood (*Populus euramericana* Guinier). This species is known to have a characteristic tension wood fibre with G-layer organised as $P+S_1+S_2+G$[17]. Samples were taken from the upper side of a tilted and bent young poplar tree (8 cm diameter at the breast height). This tree shape shows the necessity and ability of tree to restore verticality what is an indicator of the production of tension wood. Anatomical observations confirmed presence of a large amount of fibres with thick G-layer and thin $S_2$-layer in samples.

Samples preparation

Samples were maintained in water as soon as they were taken out from the tree. Wood sticks (4 mm in longitudinal direction, 1 x 1 mm² in cross section) were longitudinally cut by splitting to guarantee a good axial direction. To avoid shrinkage, the samples were always kept in a drop of water during the preparation. They were then cut in the middle of axial direction, perpendicular to the fibre, with a brand-new razor blade (Feather S35 type) in order to obtain two samples. Both samples are perfectly symmetric and the effect of the tool on both faces can be considered as identical. One sample was oven-dried (105°C during 6 hours) and the other kept in water (Fig. 1). One face being common to both sample, one can recognize fibres on both sample and then compare the effect of drying on the same fibre. This common face will be called reference face (RF). Wet sample were dehydrated with ethanol series and embedded in LR White resin (two exchanges of resin/ethanol mixture for 1 hour, followed by two exchanges in pure resin for 1 hour and kept overnight at room temperature). Dried sample were directly embedded in LR White resin after being removed from the oven (two exchanges under vacuum in pure resin for 1 hour and kept overnight at room temperature). Serial cross-sectioning (2 µm thickness) was performed with a glass knife. Sectioning of oven-dried sample was more difficult and flatness of the sections often more irregular. It is likely because the penetration of the resin into dry samples without ethanol series is less efficient. Sections were stained with Toluidine blue mixed with Azure II, mounted on glass slides and observed under an optical microscope. Images were obtained with a digital camera and measurements were done with image analysis software.
After polymerisation of the resin, all deformations of the tissue are supposed to be blocked, and then sectioning does not alter the shape and the size of the cell wall layers (compression deformations inevitably produced by the cutting effort, perpendicularly to the cutting direction, were not considered because they do not get involved in the interpretation of results in this article). This method allows to do some observations of the cells from the end (RF) to inside the sample conserving the morphology as it was before embedding. Thus, all the deformations observed in the cell shapes are the results from the RF cutting and drying shrinkage before the embedding. As a proof that G-layer detachment has been produced before embedding, the presence of resin between G-layer and $S_2$-layer can be observed (continuity of knife trace in resin between G-layer and $S_2$-layer) in Fig. 2 a-b. Shape of the cell wall layers (notably G-layers) was followed from the cutting end, along the fibres. Detachment of G-layer was taken into account as far as visible under a microscope (magnification 630X).
In this report, "never-dried wood" means the wood that has resin-embedded without oven-drying. However, the consequences of dehydration by ethanol series needed for embedding are not well known. Notably, a partial shrinkage could occur as suggested by Ishimaru and Sakai[18].

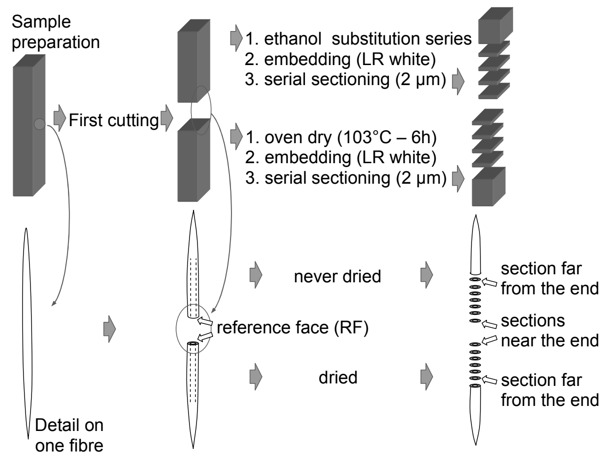


**Fig 1**: Experimental protocol and terminology used in the article

## Results

G-layer detachment in never-dried wood

As frequently observed on thin transverse sections, poplar sample studied presents some fibres where G-layer was partially detached from $S_2$-layer. This phenomenon was clearly visible at the end of the sample (near RF) but disappeared far from it in a series of sequential section. Figure 2 shows the same group of cells observed at 6 distances from the RF (10, 18, 28, 50, 70 and 150 µm respectively). 25 fibres which G-layers were detached on the surface of the sample were followed until 300 µm far from the RF. It appears that at 40 µm far from RF, only half of the fibres were still with a detached G-layer. At 100 µm from the RF, the 25 fibres observed did present no more delamination between G-layer and $S_2$-layer. Continuing observation until 300 µm, no detached G-layer was not able to be observed.

Effect of drying on G-layer detachment

In the oven-dried part of the sample, fibres presenting G-layer detachment were the same with the one presenting detachment in the non-dried part of the sample (Fig. 3). The same number of fibres presented detachment of G-layer. Similar depth (barely more) was needed to recover adherence between G-layer and $S_2$-layer. Like in the never-dried sample, after about 100 µm from the end, no detachment of G-layer was observed.

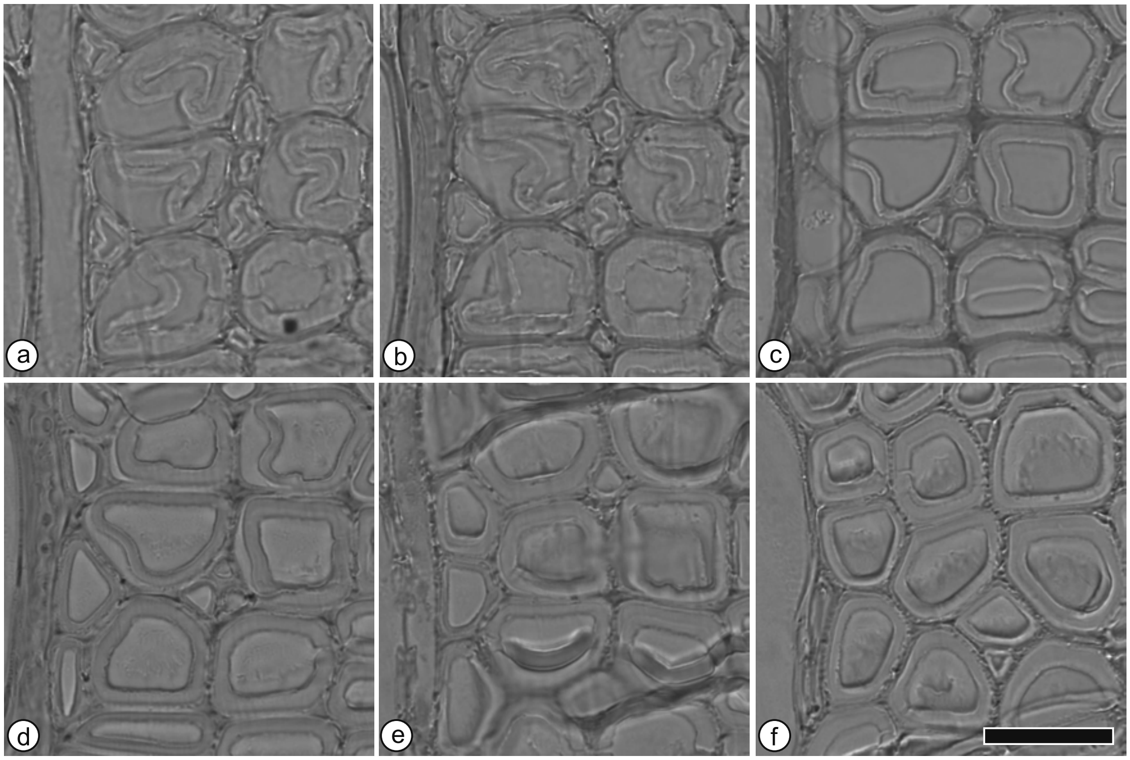

Fig. 2: Transverse section of never-dried poplar tension wood. Observation of detachment of G-layer from $S_2$-layer versus the distance (D) to the cutting surface (RF). a: D=10 µm; b: D=18 µm; c: D=28 µm; d: D=50 µm; e: D=70 µm; f: D=150 µm. Scale bar = 20 µm.

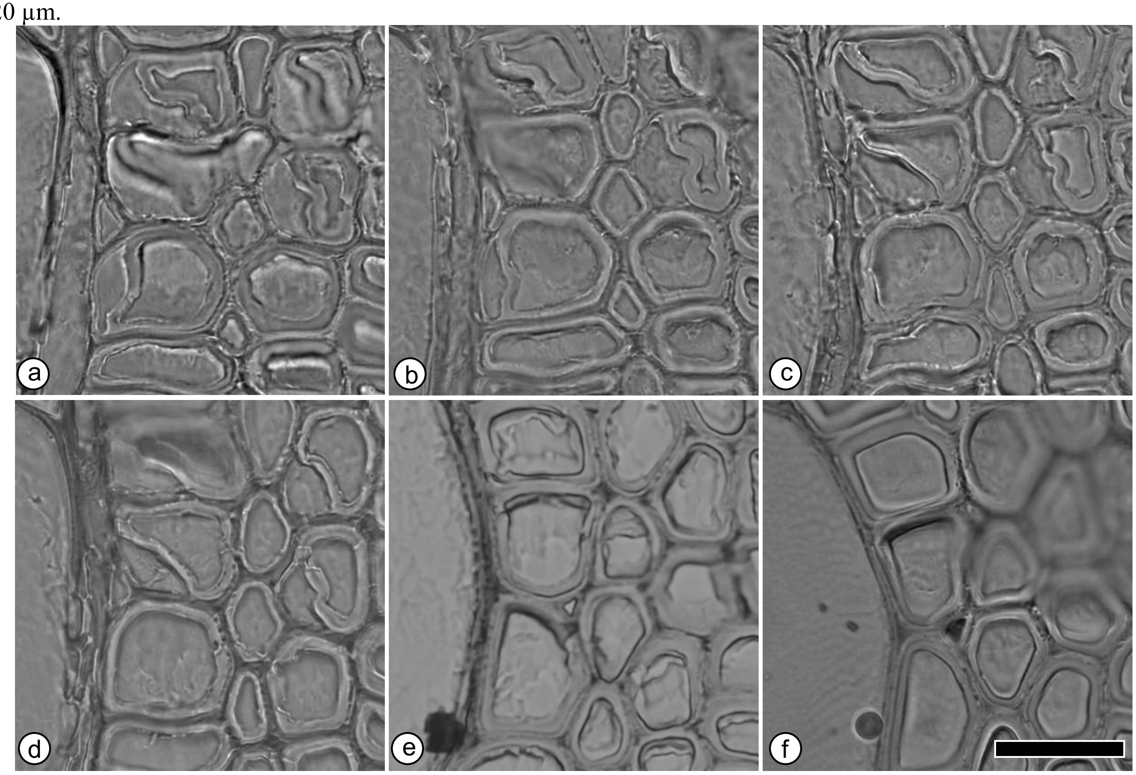

Fig. 3: Transverse section of dried poplar tension wood. Distance (D) to the cutting surface (RF): a: D=10 µm; b: D=16 µm; c: D=34 µm; d: D=50 µm; e: D=96 µm; f: D=150 µm. Observed cells are the same in Fig. 2. Scale bar = 20 µm.

## Discussion

These observations show that detachment of G-layer often observed is a border effect. In our observations, this effect affected only the first 100 µm near the end (RF). So, using a conventional sliding microtoming, detachment of G-layer can be foreseeable since the thickness of sections is usually around 10 to 20 µm.

Observations made on the dried wood shown that G-layer detachment did not or little depends on drying shrinkage.

Ours observations agree well with the observations made by Okumura et al.[19]. They follow the thickness variation of the G-layer all along tension wood fibres on embedded samples, however no detachment can be observed on the electron micrographs they presented. This may likely because in order to observe total length of the targeted fibres, the sections were cut far enough from the border of the sample. In these conditions, detachment would not be observed.

As reported by some authors[20], observation near the end of the sample (Fig. 2ab and 3ab) shows that the largest deformations of all detached G-layer are always oriented in a same direction. Then, action of the tool (razor blade) on the G-layer seems to be the trigger of the detachment. However, others layers than G-layer has never been reported to be subject to detachment during sectioning. Then the specificity of G-layer will have to be considered to explain its detachment from $S_2$-layer. Some works are in progress to show if the high tensile stress which can be expected in G-layer[21] could be the trigger of this detachment.

Thus, in tension wood fibre, G-layer is always in adherence to the $S_2$-layer in massive wood. Adherence is strong enough not to be too much altered by the high transverse and longitudinal shrinkage of G-layer. These observations prove the contribution of G-layer to the mechanical and physical properties of tension wood. As the G-layer shrinks during drying[12], the present study reinforce the idea that G-layer is the driving force of macroscopic longitudinal shrinkage of tension wood.


## Acknowledgements

The study was supported by a Grant in Aid for Scientific Research from the Japanese Society of Promotion of Science (no. 14656069, 14360099,14002805). CB is a recipient of a JSPS Fellowship.


## References


1. Fisher JB, Stevenson JW (1981) Occurence of reaction wood in branches of Dicotyledons and its role in tree architecture. Botanical Gazette 142(1): 82-95
2. Wardrop AB (1964) Reaction anatomy of arborescent angiosperms. In: H Zimmermann (ed): The formation of wood in forest tree. Academic Press, New York. London, pp 405-455
3. Onaka F (1949) Studies on compression and tension wood. Wood Research, Bulletin of the Wood Research Institute, Kyoto University, Japan 24(3): 1-88
4. Côté WA, Day AC, Timell TE (1969) A contribution to the ultrastructure of tension wood fibers. Wood Science and Technology 3(4): 257-271
5. Norberg PH, Meier H (1966) Physical and chemical properties of the gelatinous layer in tension wood fibre of aspen (*Populus tremula* L). Holzforschung 20: 174-178
6. Fujita M, Saiki H, Harada H (1974) Electron microscopy of microtubules and cellulose microfibrils in secondary wall formation of poplar tension wood fibers. Mokuzai Gakkaishi 20(4): 147-156
7. Grzeskowiak V, Sassus F, Fournier M (1996) Macroscopic staining, maturation and drying longitudinal shrinkage of tension wood of poplar (*Populus euramericana cv. I.214*.) (in French). Ann. Sci. For. 53(6): 1083-1097
8. Skaar C (1988) Wood-Water Relations. Series in Wood Science. Springer-Verlag, Berlin, 283 pp
9. Washusen R, Ilic J (2001) Relationship between transverse shrinkage and tension wood from three provenance of *Eucalyptus globulus* Labill. Holz als Roh- und Werkstoff 59: 85-93
10. Clair B, Jaouen G, Beauchêne J, Fournier M (2003) Mapping radial, tangential and longitudinal shrinkages and its relation to tension wood in discs of the tropical tree *Symphonia globulifera*. Holzforschung 57(6): 665-671
11. Clair B, Ruelle J, Thibaut B (2003) Relationship between growth stresses, mechano-physical properties and proportion of fibre with gelatinous layer in chestnut (*Castanea sativa* Mill.). Holzforschung 57(2): 189-195
12. Clair B, Thibaut B (2001) Shrinkage of the gelatinous layer of poplar and beech tension wood. IAWA Journal 22(2): 121-131
13. Wardrop AB, Dadswell HE (1955) The nature of reaction wood. IV. Variation in cell wall organization of tension wood fibres. Australian Journal of Botany 3(2): 177-189
14. Isebrands IG, Bensend DW (1972) Incidence and structure of gelatinous fibers within rapid-growing eastern cottonwood. Wood and Fiber 4(2): 61-71
15. Côté WA, Day AC (1964) Anatomy and ultrastructure of reaction wood. In: WA Côté (ed): Cellular ultrastructure of woody plants. Syracuse University Press, New York, pp 391-418
16. Boyd JD (1977) Relationship between fibre morphology and shrinkage of wood. Wood Science and Technology 11: 3-22
17. Araki N, Fujita M, Saiki H, Harada H (1982) Transition of the fiber wall structure from normal wood to tension wood in *Robinia pseudoacacia* L. and *Populus euramericana* Guinier. Mokuzai Gakkaishi 28(5): 267-273
18. Ishimaru Y, Sakai H (1988) Swelling of Wood in Liquid Mixtures : I. Water-ethanol and water-acetone. Mokuzai Gakkaishi 34(11): 889-894
19. Okumura S, Harada H, Saiki H (1977) Thickness variation of the G layer along a mature and a differentiating tension wood fiber in *Populus euramericana*. Wood Science and Technology 11(1): 23-32
20. Ritter DC, Kroll RE, Gertjejansen RO (1993) Zones of Gelatinous fibers in *Populus balsamifera* L. Wood and Fiber Science 25(2): 198-208
21. Clair B, Sugiyama J, Gril J, Thibaut B (2003) Some ideas about the structural aspect of the gelatinous layer from tension wood. In: FW Telewski (ed): 4th Plant Biomechanics Conference, East Lansing, U.S.A., 29