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Safranine Penetration Path Observed by Optical Microscope in Four Korean Pine Wood Species[1]

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ABSTRACT

Optical microscope was used to observe the difference of safranine penetration in *Pinus densiflora, Pinus rigida, Pinus koraiensis* and *Larix kaempferi* grown in Korea. It was found that *Pinus koraiensis* contained the highest number of ray parenchyma and ray tracheids. In longitudinal direction, latewood penetration was found higher than that of earlywood. The number of resin canals was found highest in *Pinus koraiensis* and lowest in *Pinus rigida*. The resin canal conducted safranine higher than longitudinal tracheids. In longitudinal direction, safranine diffused from longitudinal tracheid to ray parenchyma through the cross-field pits and from the longitudinal resin canal to ray parenchyma or longitudinal tracheid. Safranine diffused from longitudinal tracheid to its neighboring tracheid through bordered pit or ray parenchyma through the cross-field pits.

Key words: Liquid penetration, Safranine, Resin canal, Diffusion, Cross-field pit.

INTRODUCTION

The mechanism of liquid flow in wood plays an important role for the processing of wood which is related to the wood preservation treatment, fire retardant treatment, dimensional stability and wood drying. Anatomical characteristics of wood- especially, capillary structures of wood are important factors to determine the liquid penetration in wood. Liquid enters in wood following different path. In axial flow, longitudinal tracheid is the main avenue for liquid penetration. While the ray parenchyma plays an important role for transverse liquid flow (Wardrop and Davies 1961). Howerver, theories concerning spreading from ray elements to longitudinal tracheids (or vice versa) are still disputed. For softwood, the ray tracheids situated at the outer most tunnels in a ray, are often found to serve as an important liquid transport path during impregnation (Liese and Bauch 1967). Main capillary structures consist of longitudinal tracheids as well as ray cell, the resin canal and pit membrane play an important role in liquid penetration of wood (Kim and Park 1991). Microscopy has been used in many investigations to localize the distribution of a penetrant in wood. The present investigation was undertaken to find out the cause for the variation of permeability of

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safranine in four Korean pine wood species using optical microscope. This paper also describes the path of safranine penetration.

MATERIALS AND METHODS

Wood species used

Four kinds of wood species were used: ① *Pinus koraiensis* Sieb. et Zucc. ② *Pinus densiflora* Sieb. et Zucc. ③ *Pinus rigida* Mill. and ④ *Larix kaempferi* Carr. Wood samples were collected from the Kangwon National University reserve forest at breast height. Immediately after the collection, discs were made and kept in air tight cellophane bags to protect from moisture loss.

Sample preparation and estimation of the moisture content (%)

The samples of 6 cm (length) x 1 cm (width) x 0.5 cm (thickness) were prepared from the collected discs by separating sapwood and heartwood. Wood blocks were weighed and dried in an oven for 24 hours maintaining the temperature 105 ^{o}C. The Moisture content of wood blocks was calculated in wet weight basis.

A counting of 25 was performed to find out the mean values of ray parenchyma, ray tracheid, resin canal and pit numbers. Structural differences among the species were tested by using a one-way ANOVA. When significant differences occurred ($P \leq 0.05$), the ANOVA procedure was followed by a Duncan significant difference post hoc test to separate the species effects (SPSS, Version 11.5.0, 2002). For radial direction penetration, except one radial and tangential surface, all surfaces were coated with silicon resin. Longitudinal penetration was observed in radial surface. So, except one cross and radial surface, all surfaces were coated with silicon resin for preventing the leakage by other surfaces. Samples were soaked in 1% safranine solution for 5 minutes. After inputting the safranine solution in different wood, permanent slides were prepared using sliding microtome and were observed under optical microscope to find out the cells responsible for safranine impregnation in both sapwood and heartwood.

RESULTS AND DISCUSSION

By optical microscope, the numbers of resin canal, ray parenchyma, ray tracheid and pits presented in the endwall pit were counted. They are shown below-

Table 1 Number of ray parenchyma, ray tracheid and resin canals in different wood species observed under optical microscope

Wood species		*P. koraiensis*	*P. densiflora*	*P. rigida*	*L. kaempferi*
Ray parenchyma (range)		5NS (1~12)	4NS (1~7)	3NS (1~6)	4NS (1~9)
Ray tracheids		2b	3ab	4a	2b
Early wood (endwall pit)	Pit/Ray parenchyma	1b	1b	1b	3a
	Pit /Ray tracheids	2b	3a	3a	2ab
Late wood (endwall pit)	Pit/Ray parenchyma	1b	1b	1b	3a
	Pit/Ray tracheids	2b	3a	3a	2ab
Early wood	Resin canal	8a	6a	2b	3b
Late wood	Resin canal	2b	3ab	3b	5a

Means with the same letter are not significantly different at p=0.05
NS: No significant at the 5% level of probability

It was found that *P. koraiensis* had the highest number of ray parenchyma in the ray structure and *P. rigida* contained the highest number of ray tracheids. *L. kaempferi* contained the highest number of pits in the endwall of ray parenchyma and ray tracheids of earlywood. The counting of the endwall pit was important because it was one of the factors which were related to transverse conduction of safranine. The resin canal also conducted liquid in longitudinal direction. *P. koraiensis* and *L. kaempferi* contained the highest number of resin canals in earlywood and latewood, respectively. From the result of optical microscope, it was observed that *L. kaempferi* had the highest number of the endwall pits in ray parenchyma and ray tracheid.

Table 2 Tracheids diameter and wall thickness in coniferous Unit : ㎛

Classification	Length	Diameter of radial direction		Diameter of tangential direction	
		Earlywood	Latewood	Earlywood	Latewood
P. koraiensis	3,410	41.9	22.4	39.0	37.7
P. densiflora	3,270	39.5	22.9	34.3	35.4
P. rigida	3,470	43.4	23.6	35.2	31.7
L. kaempferi	3,670	48.0	18.3	30.9	31.1

Source: Park et al. 1990

In this experiment the moisture content of *P. koraiensis, P. densiflora, P. rigida* and *L. kaempferi* were found 6.9%, 9.0%, 6.9% and 6.0% respectively. Besides the moisture content, permeability varied considerably based on capillary structure. Wood is a capillary porous medium and that pore structure can be defined by the cell lumen and the cell wall openings through pits which interconnect cell to cell. If the pit membrane opening are large and numerous, permeability will be higher (Comstock 1967). The primary routes for liquid penetration into wood are provided by longitudinal tracheid, resin canal, ray parenchyma and ray tracheid. So, longitudinal tracheid length and diameter are also important factors for longitudinal penetration. Table 2 shows the data for longitudinal tracheid length and diameter.

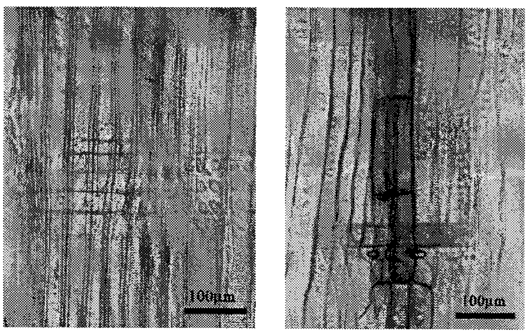

Fig.1. In radial direction, safranine penetration in sapwood (left) and heartwood (right) of *P. rigida.*

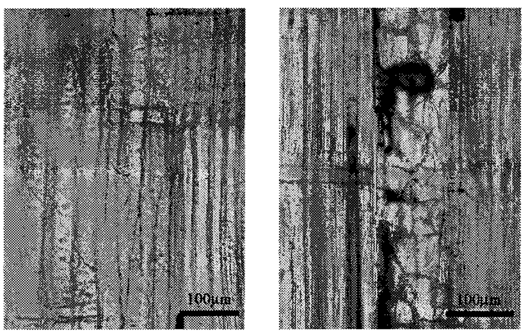

Fig.2. In radial direction, safranine penetration in sapwood (left) and heartwood (right) of *L. kaempferi.*

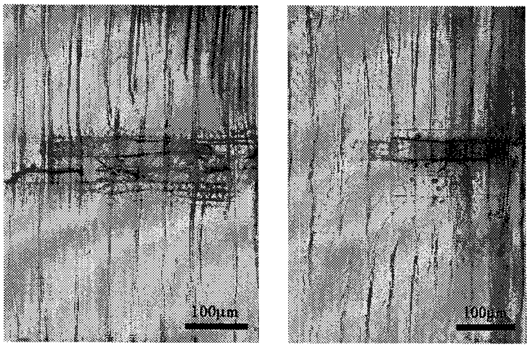

Fig.3. In longitudinal direction, sap latewood (left) and resin canal in heartwood (right) conduction of safranine in *P. koraiensis*.

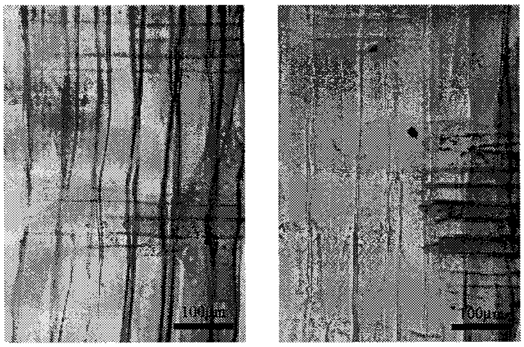

Fig.4. In longitudinal direction, sap early and latewood (left) conduction of safranine and tylosoid (right) in *P. densiflora* heartwood.

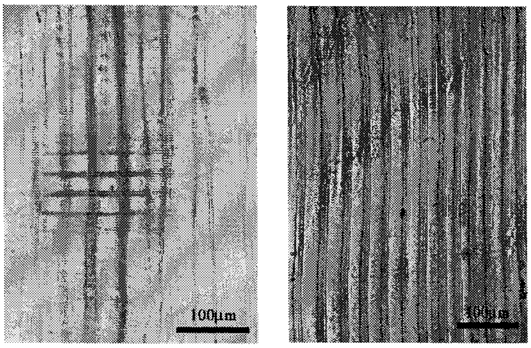

Fig.5. In longitudinal direction, sap earlywood (left) and latewood (right) conduction of safranine in *P. rigida*.

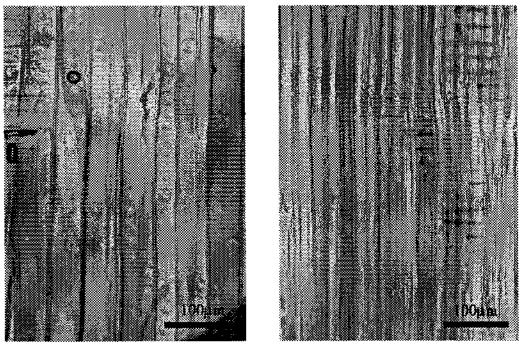

Fig.6. In longitudinal direction, safranine conduction by earlywood of L. kaempferi (left) and latewood of *P. densiflora* (right)

Ray tracheids usually present above and below of ray parenchyma. Ray parenchyma and ray tracheid were found easily permeable in sapwood. But ray tracheid conductivity was low in heartwood for all wood species (Figs. 1 and 2). For this reason, the heartwood safranine penetration was lower than sapwood. Vertical Resin duct also conducted safranine in longitudinal direction (Fig. 3). It is clear that safranine was diffused from ray parenchyma to longitudinal tracheid through the cross-field pits. Tylosoid present in heartwood could not able to conduct safranine (Fig. 4). From longitudinal tracheid, safranine was diffused to ray parenchyma through the cross-field pits (Fig. 5). In longitudinal penetration, it was found that latewood penetration was higher than earlywood penetration (Fig. 5 and 6). This phenomenon can be resulted from the capillary action (Chun and Ahmed, 2006). From vertical resin ducts, safranine was also diffused to longitudinal tracheid (Fig. 3). Finally the path of safranine penetration would be like it can diffuse to ray cells from longitudinal tracheid and then move along for a distance, it can enter to the longitudinal tracheid through the cross-field pits and so on.

CONCLUSIONS

From the optical microscopic observation, it was found that *P. koraiensis* contained the highest number of ray parenchyma and ray tracheid in ray structure. Ray played an important role for the penetration of safranine in radial direction. But heartwood tracheid had lower permeability than ray parenchyma. Early and latewood penetration in longitudinal direction was not found to be the same. Latewood conduction was observed to be higher than earlywood for both in sapwood and heartwood. Tylosoid was found to be impermeable. The number of resin canals was found the highest in *P. koraiensis* and the lowest in *P. rigida*. Resin canals diffused safranine higher than longitudinal tracheid. In longitudinal direction, the path of safranine impregnation was found that it diffused from longitudinal tracheid to ray parenchyma through the cross-field pits and from longitudinal resin canal to ray parenchyma or longitudinal tracheid. Furthermore, it diffused from the longitudinal tracheid to its neighboring tracheid through the bordered pit or ray parenchyma through the cross-field pits.

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