

**VISUAL SIMULATION ON DYNAMIC ACCUMULATION
OF THE DIFFERENTIATING XYLEM CELLS**

ENHUA XI

COLLEGE OF FORESTRY, SHANDONG AGRICULTURAL UNIVERSITY

TAI'AN, CHINA

BEIJING KEY LABORATORY OF WOOD SCIENCE AND ENGINEERING, BEIJING FORESTRY

UNIVERSITY

BEIJING, CHINA

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ABSTRACT

This study describes the dynamic accumulation of xylem cells of the fast-growing *Populus×euramericana* cv. '74/76' during the growth phase by the methods of microscopy analysis and computer simulation technology. In order to show a more intuitive accumulation and dynamic variation process of cells in different periods, the computer simulation software was used to simulate the accumulation process of cambium and xylem cells according to the data of the accumulation and anatomical characteristics. The dynamic accumulation process of the xylem cells was visually displayed by the computational simulation technology during the active period.

KEYWORDS: Xylem cells, *Populus×euramericana* cv. '74/76', dynamic accumulation, simulation technology.

INTRODUCTION

A known and mature simulation system is L-studio (Godin et al. 2004) to model multicellular plant systems. One of its features was to implement vv-systems, a two-dimensional rewriting grammar to model cell division (Smith 2006) derived from L-systems (Lindenmayer 1968a, 1968b, 1975). L-studio and vv-systems have been applied in many recent studies on plant development (Smith et al. 2006, Bayer et al. 2009, Prusinkiewicz et al. 2009). Many cell-based modeling methods have been applied to analyze detailed cell wall mechanics. The cell modeler (Rudge and Haseloff 2005, Dupuy et al. 2008) was a two-dimensional simulation of plant tissue, which can express the interaction between the cells and the cell walls, including dynamic changes in cell characteristics of the differential equation model, as well as biochemical networks. The cell modeler as a viscoelastic rod, which was stretched by compression, was used to describe the cell wall. The differential equation was used to simulate the simulation of cell wall dynamics, and geometric model was used to describe the cell division and death. The cell walls were also

demonstrated by Corson et al. (2009) as viscoelastic beams, but with an energy minimization method to simulate cell wall kinetics and cell growth.

Visual research is mainly in the field of computer graphics, especially in the visualization of plant growth has been mostly based on three-dimensional graphics technology recently. Three-dimensional graphics of the way the true display of an object is still the main research content of computer graphics, but also the focus of plant growth visualization. The visualization techniques involved in plant growth simulations are different from pure 3D graphics studies, which need to combine the characteristics of plant growth in addition to the vivid shape of the plant, otherwise, they cannot meet the needs of virtual plant technology. For example, the plant organ model, not only true form is required but also morphological changes under the control of the plant growth's simulation method. Visualization technology researches cannot only visualize the plant growth, but also promote the development of computer graphics technology.

In this study, the computer graphics technology was used to simulate the accumulation and morphological changes of the number of cells in the developmental process of *Populus×euramericana* cv. '74 /76' during the active period based on the test and observation of the morphology and proportion of the cambium and xylem cells.

MATERIAL AND METHODS

Healthy plants of fast-growing three year-old *Populus×euramericana* cv.'74/76' grown in a plantation in Beijing (40°17"N, 116°39"E; Beijing, China) with the same diameter at breast height were chosen and marked. Plant material was collected 1.3 m above the ground root level and sampled monthly from April to October (four times a month, that is, 7, 15, 22 and 30 of each month). On each occasion, blocks of about 10 mm³ including phloem, cambium and xylem cells were immediately immersed in fixative formalin–acetic acid–alcohol (FAA) for preserving the material. Upon returning to the laboratory, they were placed in the same fresh fixative under a slight vacuum for 30 minutes. Following vacuum, these pieces were fixed in fresh fixative and preserved at 4°C. Cross slices with a thickness of 10 μm including phloem, cambium, and xylem were cut on a sliding microtome, observed with a polarization microscope. Meanwhile, the morphology of cambium and xylem cells in all developmental phases was collected using a microscopic image analyzer.

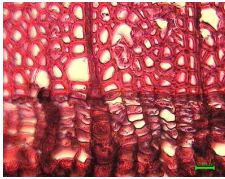
The image processing software Photoshop combining with the pictures and data collected was used to imitate the dynamic accumulation of xylem cells during the growth period. The pictures that were drawn in Photoshop were imported to the Flash. In the interactive interface of Flash, the AS3.0 scripting language was taken to write interaction design in different frames. Finally, the dynamic development process of cells was created to make it into visualization.

RESULTS AND DISCUSSION

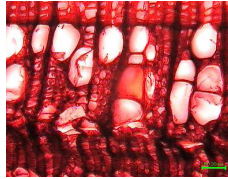
The accumulation of cambium and xylem cells during the growth phase

According to Fig. 1, on April 7 (Fig. 1, a), the cambial cells (5~7 layers of cambium cells) were swelling and showed few periclinal divisions, indicating the beginning of cambium activity. When the cambium was active, there were a large number of immature xylem cells differentiating. On May 7, the cambium was just beginning to split, and approximately 7 layers of xylem immature cells and 10 layers of cells in the cambial zone were discovered (Fig. 1, b). On July 15, the 9 layers of cambium fusiform cells and many layers (10~25) of immature xylem cells were found, indicating a high activity of cambium (Fig. 1, c). On September 7, the cambium

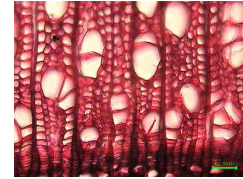
activity was reduced to a minimum, with 5 to 10 layers of cells in cambium and 7~12 layers of immature xylem cells (Fig. 1, d). When the cambium entered in the dormant phase, the cambial zone started to become narrow, with relatively thick radial walls and surrounded by no layers or only a few (1~2) layers of immature secondary xylem. Dormant cambium was shown on October 7 (Fig. 1, e) and October 15 (Fig. 1, f), and contained 5~7 cells in the radial rows. During this period, there were 0~2 immature xylem cells.



a. Cambium was in the beginning of its activity on April 7



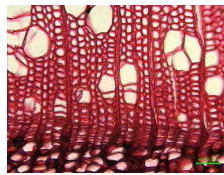
b. Cambium started to differentiate into immature xylem cells on May 7



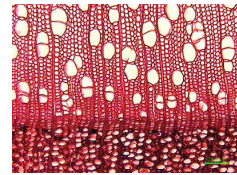
c. The differentiation capacity of cambium reached to peak on July 15



d. The immature xylem cells began to reduce on September 7



e. Cambium was in dormant phase on October 7



f. There were no immature xylem cells on October 15

Fig. 1: Microstructure of cross sections of *Populus×euramericana* cv. '74/76'

The seasonal changes (from April to October) of the Cambium (cell number, tangential and radial width) and xylem cells (vessel element, wood fiber, parenchyma and wood ray) during the active phase can be seen in Tab. 1. The first week was corresponded to the April 30, followed by the number of weeks in this order (Tab. 1).

Tab. 1: The seasonal changes of the cambium and xylem cells in different stages during the active phase.

Week	Number	1	2	3	4	5	6	7	8	
Cambium	Cell number	7	10	11	10	10	10	11	10	
	Tangential width (um)	23.29	16.26	17.54	18.37	19.06	21.99	21.19	22.90	
	Radial width (um)	7.44	5.71	6.85	6.51	5.47	3.76	5.14	5.05	
Xylem	Vessel element	Immaturate number	0	1	1	2	3	2	4	2
		Maturate number	0	2	3	5	8	10	13	11
		Tangential width (um)			45.91	40.59	36.26	44.32	35.87	41.30
		Distribution frequency		184	185	188	132	187	151	114
		wall to lumen			0.18	0.18	0.15	0.19	0.20	0.22
		Ratio of vessel element (%)	0	32.19	25.77	20.19	23.25	28.00	32.77	31.69

WOOD RESEARCH

	Wood fiber	Immature number	0	6	8	10	10	9	16	13
		Mature number	0	4	11	33	53	58	66	70
		Tangential width/ um			23.08	22.51	25.91	20.61	21.33	20.91
		wall to lumen			0.38	0.38	0.35	0.38	0.36	0.32
		Ratio of wood fiber (%)	0	61.75	67.69	71.84	64.46	61.97	58.04	59.67
	Ratio of parenchyma (%)	0	1.4	0.86	1.34	1.40	0.86	0.59	0.67	
		Ratio of wood ray (%)	0	4.68	5.7	5.96	4.68	7.16	5.10	7.97

Number		Week								
			10	11	12	13	14	15	16	
Cambium	Cell number	12	11	9	9	11	9	10	8	
	Tangential width (um)	19.32	23.98	20.30	19.21	20.23	23.98	21.11	20.77	
	Radial width (um)	4.97	4.27	5.06	4.55	3.93	4.87	5.72	5.45	
Xylem	Vessel element	Immature number	2	2	2	1	2	1	1	1
		Mature number	15	20	17	25	29	28	24	30
		Tangential width (um)	39.48	39.69	36.52	41.69	41.53	40.41	46.46	44.23
		Distribution frequency	151	127	197	110	102	110	105	115
		wall to lumen	0.19	0.16	0.22	0.17	0.14	0.16	0.14	0.15
		Ratio (%)	35.08	31.88	32.11	28.09	25.55	23.19	23.48	21.01
	Wood fiber	Immature number	12	9	14	13	7	7	6	9
		Mature number	89	78	75	87	88	96	77	92
		Tangential width (um)	20.91	20.79	21.86	23.59	22.10	22.28	20.56	21.14
		wall to lumen	0.37	0.29	0.37	0.35	0.31	0.51	0.38	0.44
		Ratio (%)	61.56	61.36	60.9	65.09	67.41	72.48	68.54	73.92
		Ratio of parenchyma (%)	0.56	0.55	0.59	0.55	0.56	0.56	0.64	0.50
		Ratio of wood ray (%)	6.29	6.21	5.86	6.17	6.48	6.28	7.34	6.57

Week Number	17	18	19	20	21	22	23	24	25
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The visualization of cells accumulation during the differentiating process

Cambium	Cell number		11	10	9	8	9	9	7	6	6
	Tangential width (um)		25.17	18.44	22.46	19.01	23.24	22.41	21.26	20.13	19.35
	Radial width (um)		5.77	4.43	3.59	4.91	4.06	4.04	4.35	4.21	3.54
Xylem	Vessel element	Immature number	1	1	1	2	0	0	0	0	0
		Mature number	30	29	24	27	30	27	36	25	36
		Tangential width (um)	45.79	40.49	45.21	37.97	42.12	43.07	41.68	40.26	36.38
		Distribution frequency	124	145	133	149	131	143	146	152	149
		wall to lumen	0.15	0.17	0.19	0.17	0.18	0.18	0.21	0.25	0.34
		Ratio (%)	20.70	22.44	19.55	21.22	22.44	19.69	21.13	17.96	19.47
	Wood fiber	Immature number	11	9	11	11	0	0	0	0	0
		Mature number	113	113	110	118	115	120	134	134	128
		Tangential width (um)	23.96	23.82	24.83	26.09	23.51	23.35	22.73	21.35	21.16
		wall to lumen	0.51	0.70	0.57	0.62	0.68	0.70	0.74	0.78	0.83
		Ratio (%)	72.72	72.32	74.50	69.66	70.11	73.53	73.26	75.34	68.21
	Ratio of parenchyma (%)		0.47	0.62	0.57	0.62	0.82	0.63	0.72	0.62	0.60
	Ratio of wood ray (%)		6.11	5.62	5.38	5.50	6.63	6.15	4.88	6.08	8.22

In order to show a more intuitive accumulation and dynamic variation process of cells in different periods, the computer simulation software was used to simulate the accumulation process of cambium and xylem cells according to the data (Tab. 1) of the anatomical characteristics.

The cross-section pictures of cambium and xylem cells in different phases (from April to October) were showed in Fig. 1. These pictures were made into a continuous process of development in chronological order by Photoshop. In Photoshop, the simulation map of cell development and the main feature data were put into the same interface for three consecutive periods in order to compare the growth change and development of cells in different periods. The following visual elements were included in the interactive interface: title, button, three windows (three continuous periods), the main data and scale (1:365).

In Photoshop, the non-text element png image was exported. 1280×800 mm canvas was created in Flash CS3, and the png elements were imported into the Flash in accordance with the pre-designed. The sampling time and the cell anatomical characteristic data were input on different frames according to the data provided (Fig. 2).

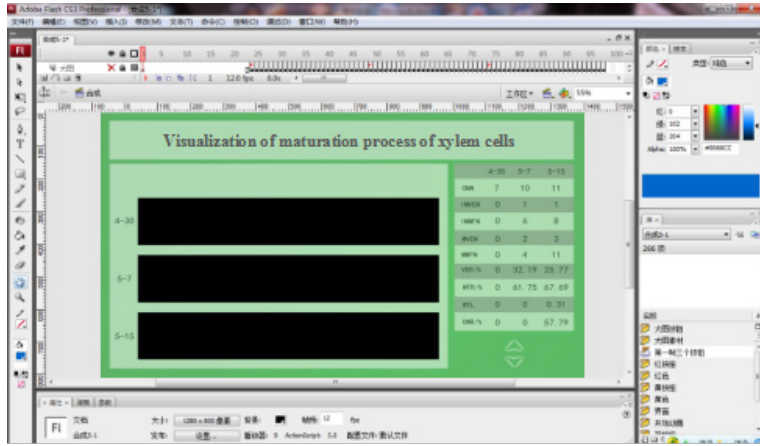


Fig. 2: Import the interface into Flash.

The scale of 1: 365 was set reasonably according to the interface size of the interface in Photoshop. Based on the collected microstructure images and the measured data, the accumulation and anatomical changes of cambium were simulated from April 30 to October 30. Similarly, the xylem cells in the active period were plotted with reference to the collected microscopic images and the data of the anatomical characteristics of the different types of cells. The draft chart on October 22 was shown in Fig. 3.

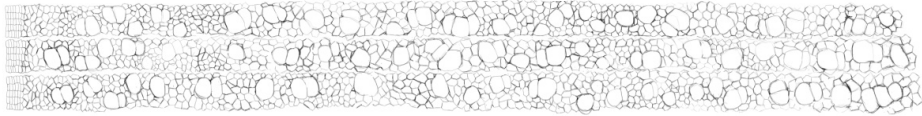


Fig. 3: Manuscript of cell development process

The each period of the jpg format manuscript was imported into the corresponding position in Flash. The cell wall in different periods was thickened according to data of the thickness in accordance with the outline of the manuscript.

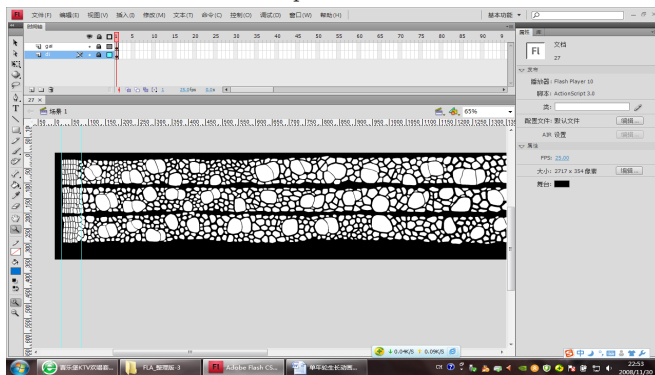


Fig. 4: Simulation of cell development process.

The white area was represented cell lumen and the black was the cell wall. The background was black. The simulation jpg picture of the cell development process on October 22 was shown in Fig. 4.

On the basis of existing cells, the bark and phloem were added to show the integrity of cell development process. The bark and phloem were illustrated, because they were not the main content of this study (Fig. 5).

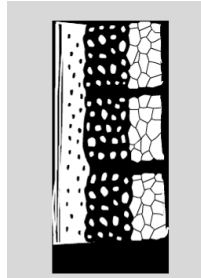
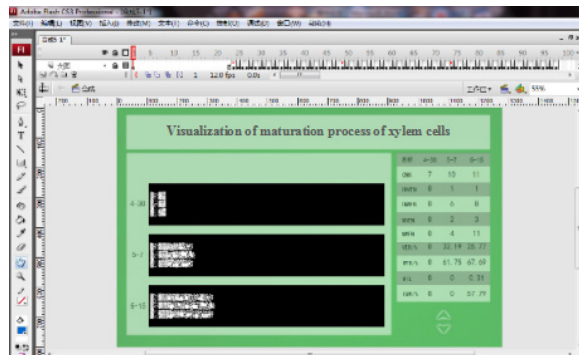
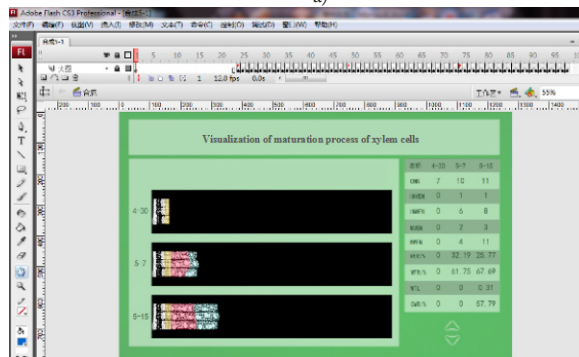


Fig. 5: Sketch of bark and phloem.

Simulated drawing of different periods of cell growth and development of jpg pictures were introduced into the corresponding interactive interface in the Flash file from Photoshop and placed according to different periods to the corresponding frame (Fig. 6a).



a)



b)

Fig. 7: Simulation pictures import into Flash.

The whole process was divided into three parts, the cambium, immature xylem cells and mature xylem cells, and were covered with yellow, red and blue in order to better distinguish the cell development process in different stages (Fig. 6b).

Interactive synthesis of cell growth and development

In the interactive interface of Flash, the AS3.0 scripting language was used writing interaction design in different frames. There were three main interactive effects in Flash: (1) When click on the up-and down button, it can go to the corresponding cell growth simulation and the data in different stages. (2) It will show the corresponding color block, when the mouse is skimmed over the cambium, immature layer and mature layer. (3) When click on the bark and phloem, it can return to the large picture corresponding to the cell growth at the same stage; when click on the big picture again, it can return to the original interface. After testing, the file was output in SWF (Small Web Format) and played with the player. The final visual simulation of the dynamic accumulation of cell layers was achieved.

Wood micro-structure modeling and even the three-dimensional structural modeling are the focus of wood basic research and the key to improve the accurate analysis of wood performance. The corresponding typical model was constructed and the elastic modulus of the cell model was calculated (Callum and Dennis 1999). The changes of wood cell diameter, cell cavity shape, cell length and cell wall thickness during the growth of spruce stem was studied by using the conventional digital image processing system (Sarén et al. 2001). The visual simulation of cell morphological and structural changes during wood growth will provide important theoretical basis for the improvement of wood properties.

CONCLUSIONS

According to the observation with the microscope, The xylem differentiation process from the cambium, immature cells and mature cells can be found. The computer technologies (Photoshop and Flash) were used to simulate the dynamic accumulation process of xylem cells during the growth phase. The visualization on developmental process of the cells was achieved.

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REFERENCES

1. Bayer, E.M., Smith, R.S., Mandel, T., Nakayama, N., Sauer, M., Prusinkiewicz, P., Kuhlemeier, C., 2009: Integration of transport-based models for phyllotaxis and midvein formation, *Genes&Development* 23: 373-384
2. Corson, F., Hamant, O., Bohn, S., Traas, J., Boudaoud, A., Couder, Y., 2009: Turning a plant tissue into a living cell froth through isotropic growth, *Proceedings of the National Academy of Sciences of the United States of America* 106(21): 8453-8458

3. Dupuy, L., Mackenzie, J., Rudge, T., Haseloff, J., 2008: A system for modelling cell-cell interactions during plant morphogenesis, *Annals of Botany (Lond)* 101: 1255-1265
4. Godin, C., Hanan, J., Kurth, W., Lacoite, A., Takenaka, A., Prusinkiewicz, P., DeJong, T., Beveridge, C., Andrieu, B., Karwowski, R., Prusinkiewicz, P., 2004: The L-system-based plant-modeling environment L-studio 4.0. In: *Proceedings of the 4th International Workshop on Functional-Structural Plant Models* (eds. Godin, C., Hanan, J., Kurth, W., Lacoite, A., Takenaka, A., Prusinkiewicz, P., DeJong, T., Beveridge, C., Andrieu, B.), UMR AMAP, Montpellier, France, Pp 403–405.
5. Lindenmayer, A., 1968a: Mathematical models for cellular interactions in development. I. Filaments with one-sided inputs, *Journal of Theoretical Biology* 18(3): 280-299
6. Lindenmayer, A., 1968b: Mathematical models for cellular interactions in development. II. Simple and branching filaments with two-sided inputs, *Journal of Theoretical Biology* 18(3): 300-315
7. Lindenmayer, A., 1975: Developmental algorithms for multicellular organisms: a survey of L-systems, *Journal of Theoretical Biology* 54(1): 3-22
8. Prusinkiewicz, P., Crawford, S., Smith, R.S., Ljung, K., Bennett, T., Ongaro, V., Leyser, O., 2009: Control of bud activation by an auxin transport switch, *Proceedings of the National Academy of Sciences of the United States of America* 106(41): 17431-17436
9. Rudge, T., Haseloff, J., 2005: A computational model of cellular morphogenesis in plants. *Lect. Notes, Computer Science* 3630, 78-87
10. Smith, C., 2006: On vertex-vertex systems and their use in geometric and biological modeling. PhD thesis, University of Calgary, Calgary, Canada.
11. Smith, R.S., Guyomarc'h, S., Mandel, T., Reinhardt, D., Kuhlemeier, C., Prusinkiewicz, P., 2006: A plausible model of phyllotaxis, *Proceedings of the National Academy of Sciences of the United States of America* 103(5): 1301-1306

ENHUA XI
SHANDONG AGRICULTURAL UNIVERSITY
COLLEGE OF FORESTRY
TAI'AN 271018
CHINA
BEIJING FORESTRY UNIVERSITY
BEIJING KEY LABORATORY OF WOOD SCIENCE AND ENGINEERING
BEIJING 100083
CHINA
Corresponding author: xienhua@126.com

