# The metabolites in developing xylem cells of annual layer in *Larix sibirica* stem.

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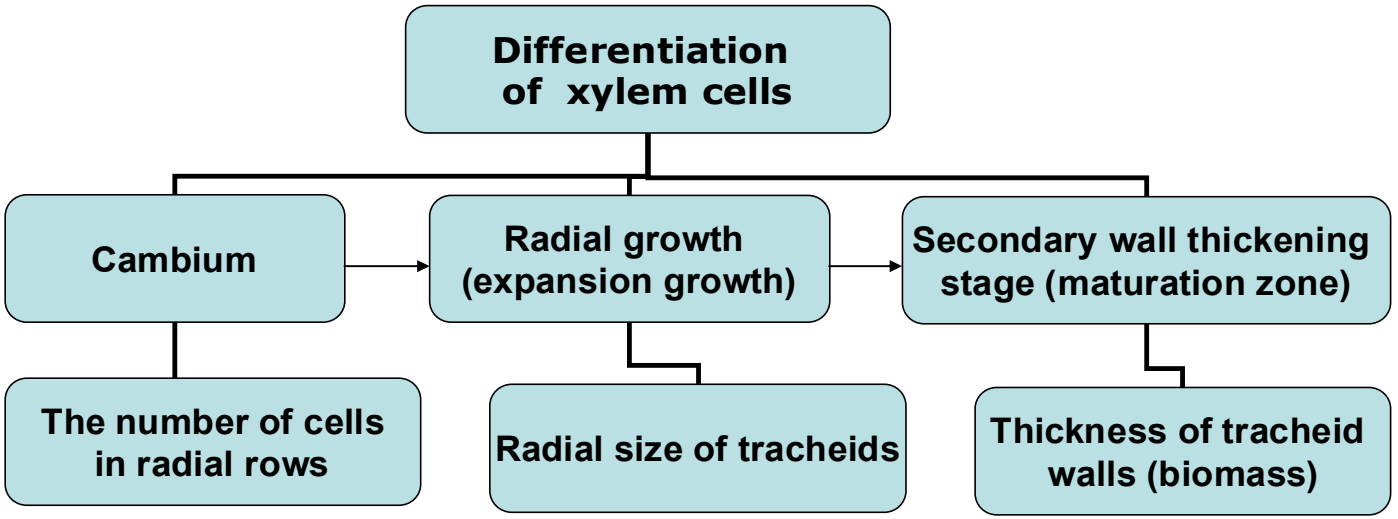
presentation Larix2012, Hallormsstadaskogur National Forest, Iceland

# Formation of annual increment in larch stem




One program, but different result:
early and late xylem tracheids in larch differ in

- tracheid diameter
- cell wall thickness

**The purpose of investigation:**

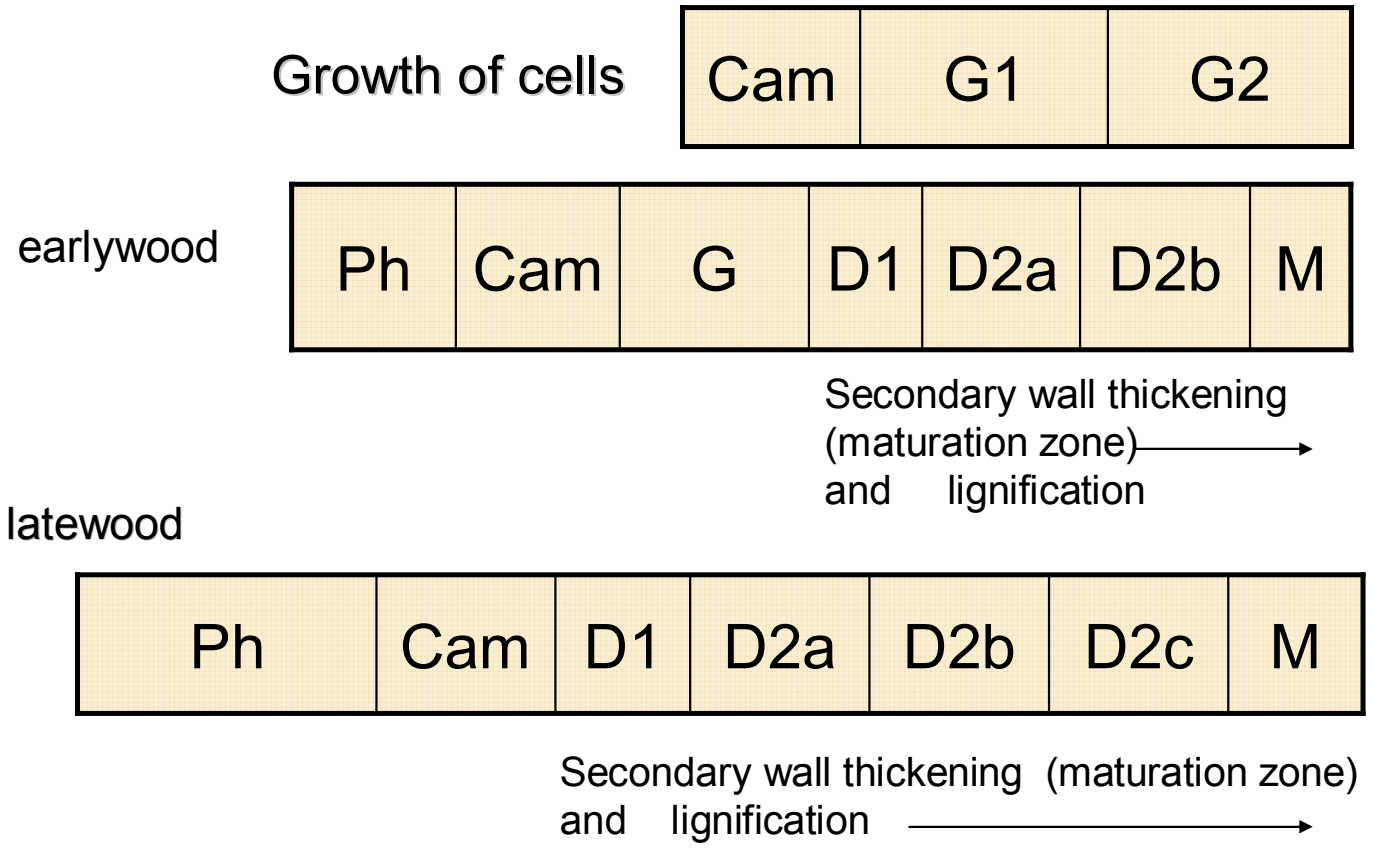
to examine the distinctions in metabolism of developing tracheids during early and late xylem formation in larch stem:

- carbohydrates, as basic product of photosynthesis, taking part in the metabolism and in creation of cell wall polymer structure,
- free and bound phenolic acids, which couple up the polymers in primary wall and are a precursors of lignin,
- ascorbic acid (AsA) and dehydroascorbic acid (DHA), as well as their ratio, showing oxidation/reduction potential of cells.

Materials:

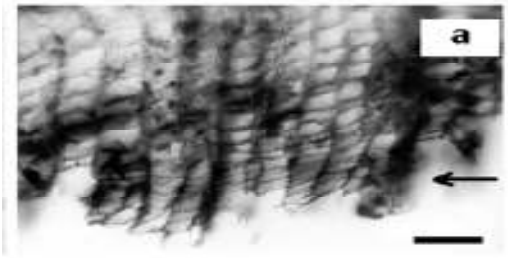
- the cell layers of forming wood in 20-25 year-old larch (*Larix sibirica* Ldb. ) trees in the periods of formation of earlywood and latewood. In Siberia this corresponds to early June and the end of June (earlywood formation) and early August – latewood formation.

# The cell layers of forming phloem and xylem



The layers of cells of cambium and expansion growth zones, sampled during formation in larch stem of

## Early xylem




**Cambium on phloem**


**First part of growth zone (G1)**

**Second part of growth zone (G2)**


**Zone of growth of earlywood tracheids (G)**

## Late xylem

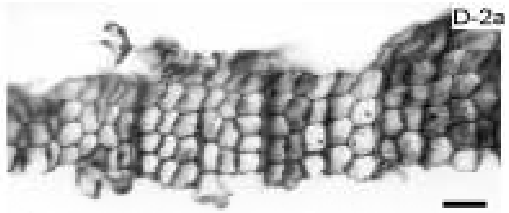

**Cambium**


**Zone of expansion growth of latewood tracheids (G)**

The layers of tracheids at successive stages of secondary wall development: D-1 - before and after D-2 a, D-2b, D-2c the beginning of lignification

**Early xylem**


**D-1**

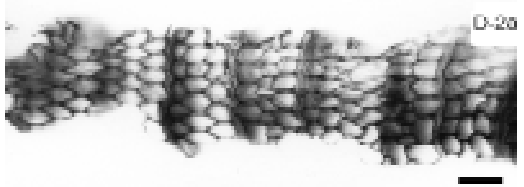



**D-2a**

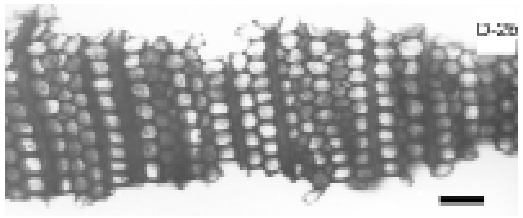

**D-2b**

**Late xylem**

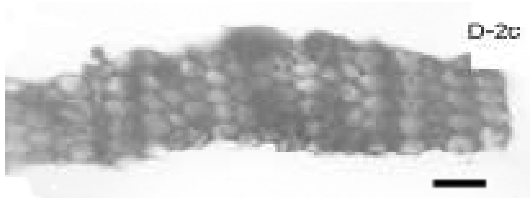

**D-1**




**D-2a**




**D-2b**

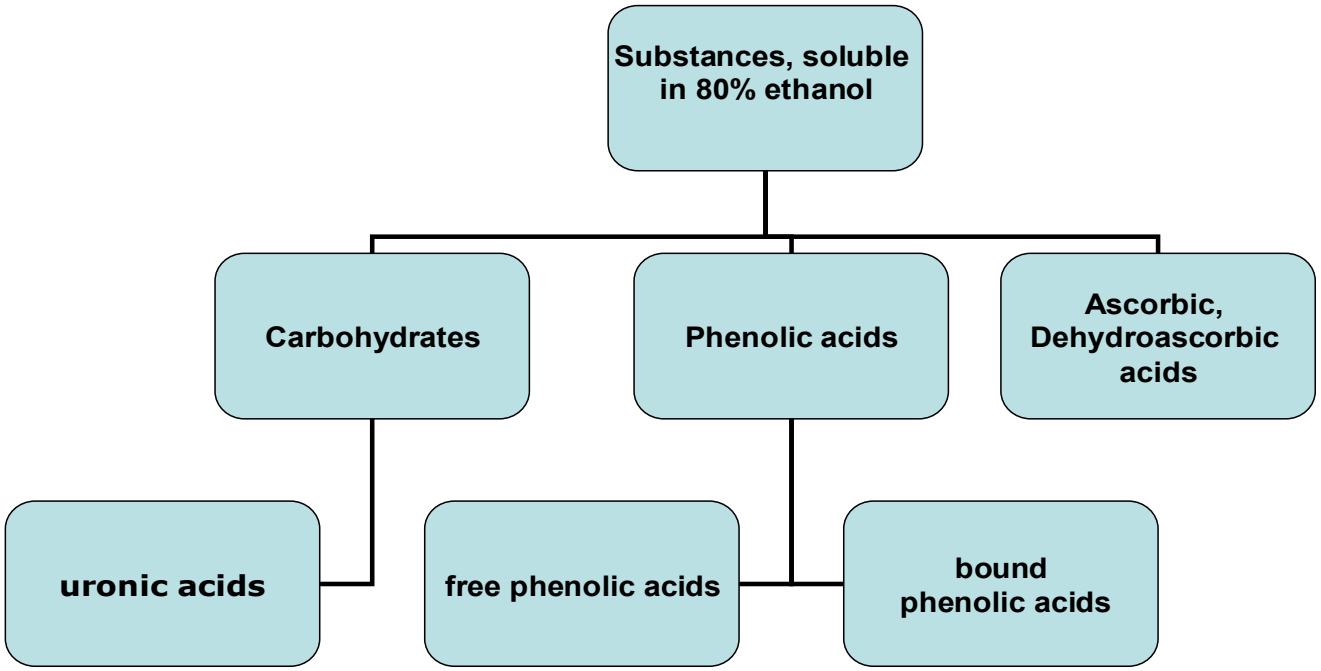



**D-2c**

## Methods:

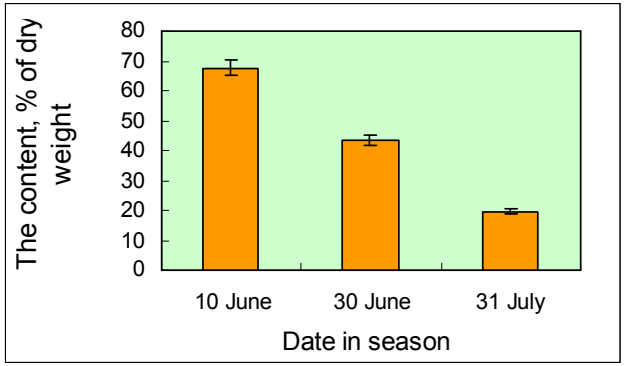
The cells with different stages of development were fixed by 96% ethanol and then processed by 80 % ethanol. The contents of carbohydrates (Dubois et al., 1956), uronic acids (Galambos, 1967) and phenolic acids (Jennings, 1981) were estimated in the extracts. Phenolic acids were fractioned by their solubility in ether. AsA and DHA were examined by (Antonova et al. 2005).

The content were calculated per dry weight and per cell.

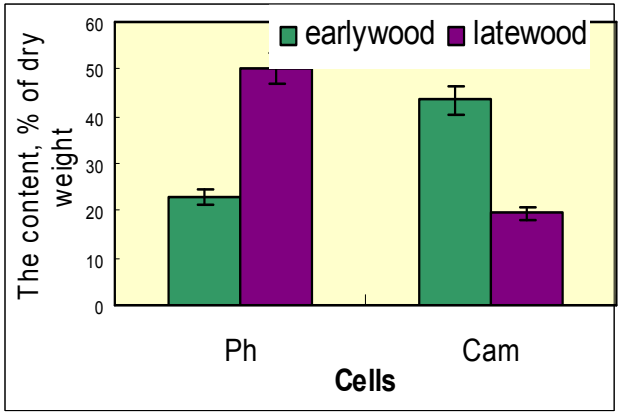


# The content of carbohydrates (a, b,c – per dry weight, d - per cell) in cambium zone (Cam) and phloem (Ph) in the season

**Cambium zone, carbohydrates during the season**

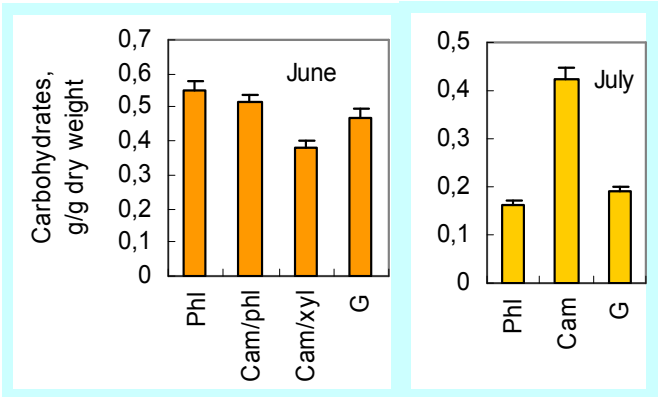



a

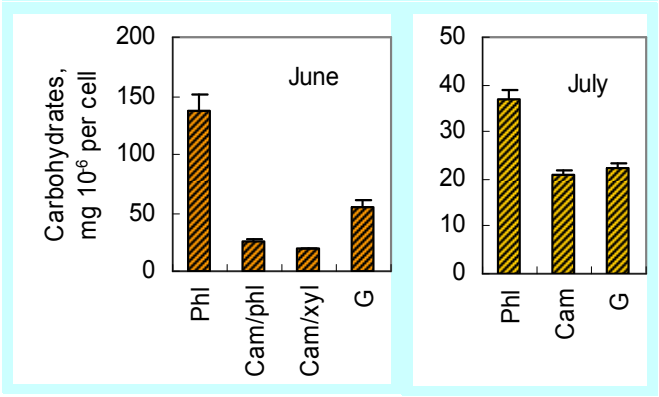



b

**Cells of phloem (Phl) and cambium zone, adjacent to Phl (Camphl) and growing (G) xylem (Camxyl)**

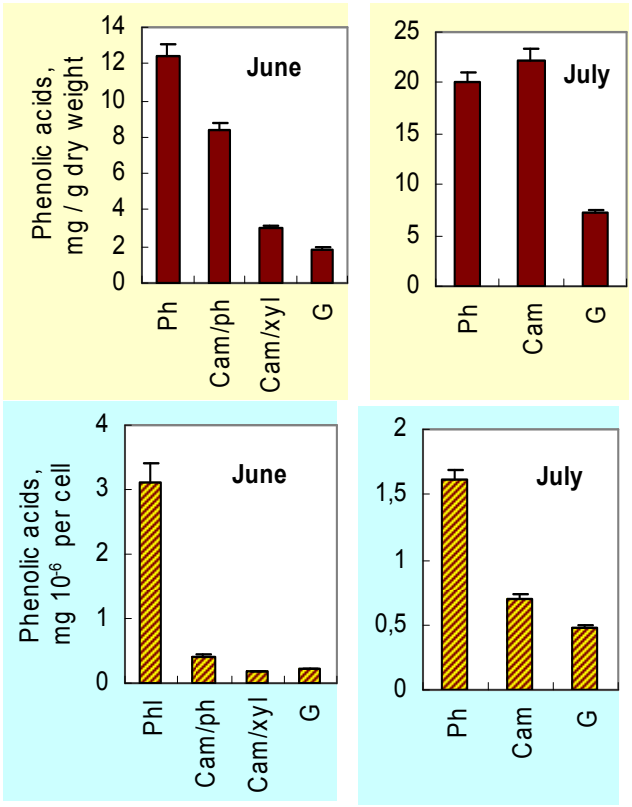



c



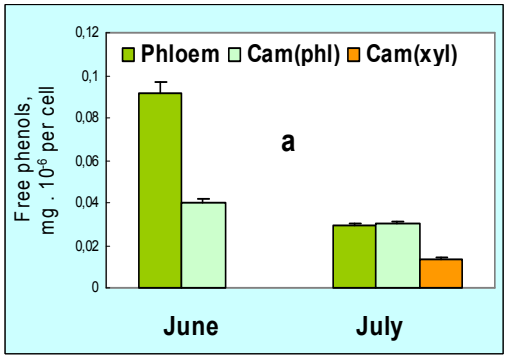

d

# The changes in the content of phenolic acids in the zones of cambium (a, b, c), phloem and expansion growth during the season

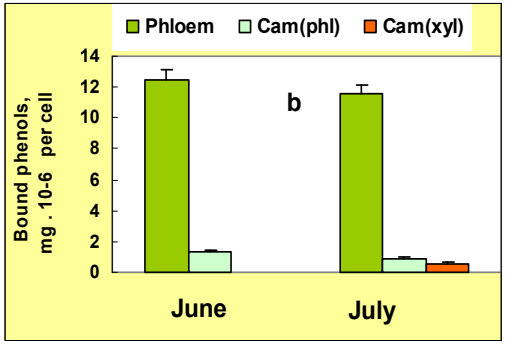



**Cells of phloem (Phl) and cambium zone, adjacent to phloem (Camphl) and expansion growth zone (G) (Camxyl)**

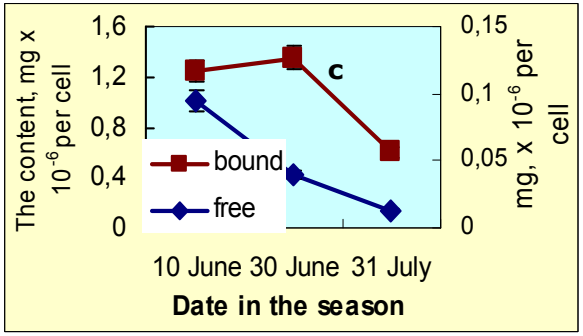
**Free acids - a**




**Bound acids - b**



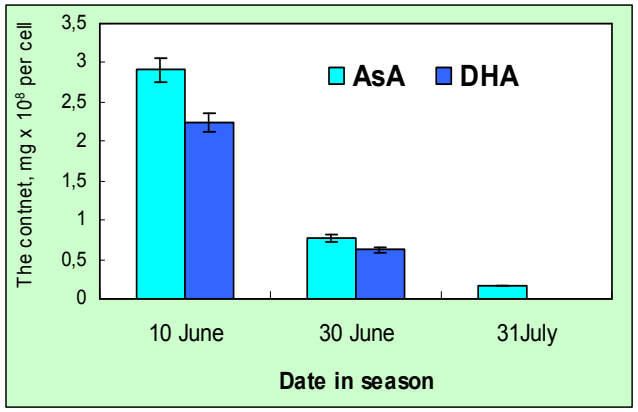

**Phenolic acid fractions - c**



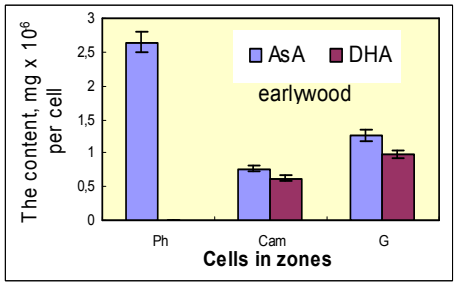

Free acids are undesirable for active processes, because are poisons for cells

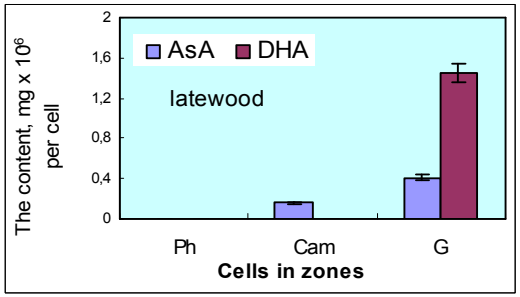
Changes in the content (per cell) of ascorbic (AsA) and dehydroascorbic (DHA) acids and of uronic acids (precursors of AsA) in cambium zone (a), in zones phloem and expansion growth (b) during the season in larch stem

during the season - **a**




**b**

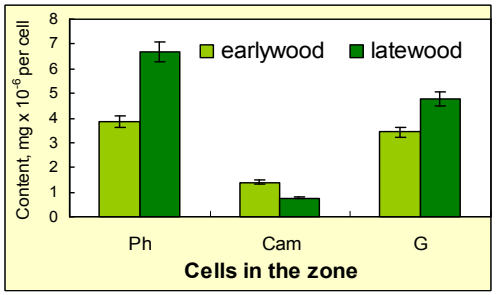



**The number of cambial initial divisions duing 10 days:**

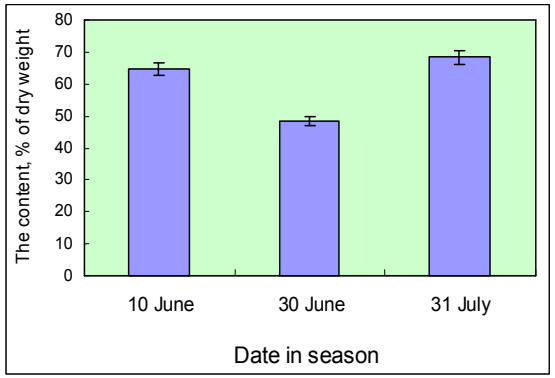
**3,2** **2,1** **1,8**

**Uronic acids**

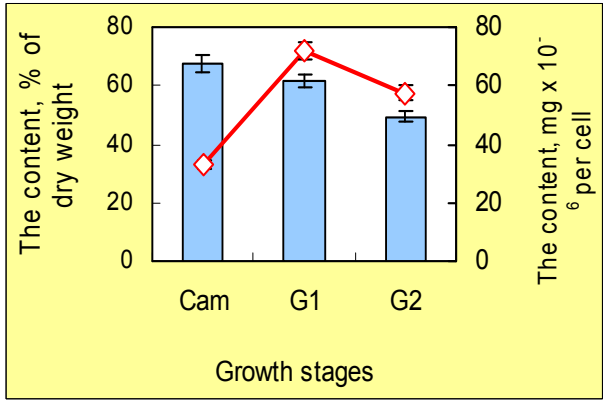


# The content of carbohydrates (a, b – per dry weight, c, d – per cell) and uronic acids (c – per cell) in larch tracheids in expansion growth zone

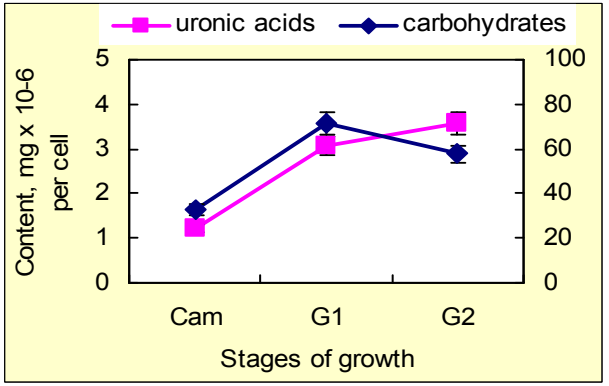
**a**-during the season



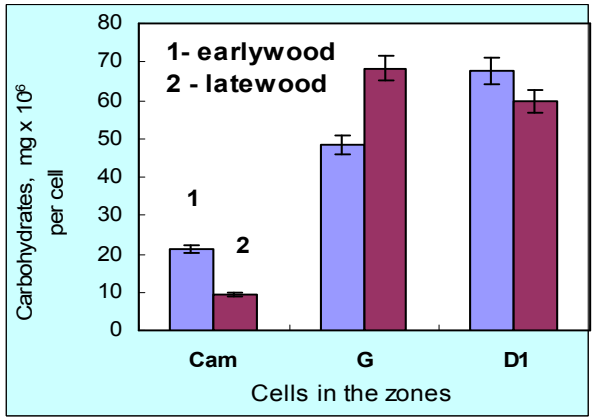

**b** – trough expansion zone




**c** – trough expansion zone




**d**

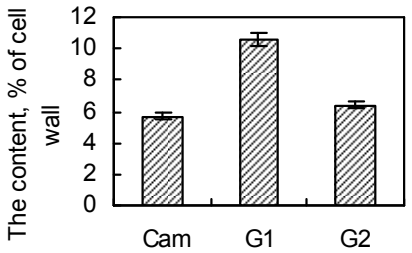



D1 – deposition of secondary wall biomass in earlywood tracheids don't is limited to carbohydrates

Changes of increment of cellulose (a), of radial diameter (b) and the content of bound and free phenolic acids (c- per dry weight, d – per cell) in the course of radial growth

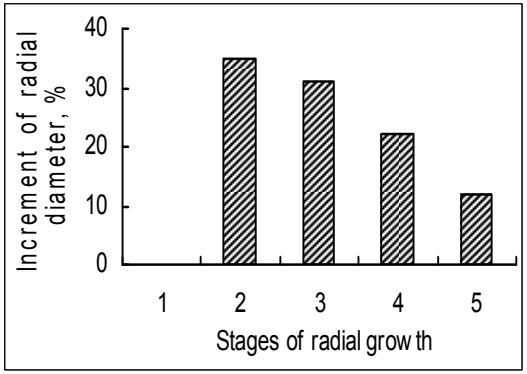
a

Increment of cellulose



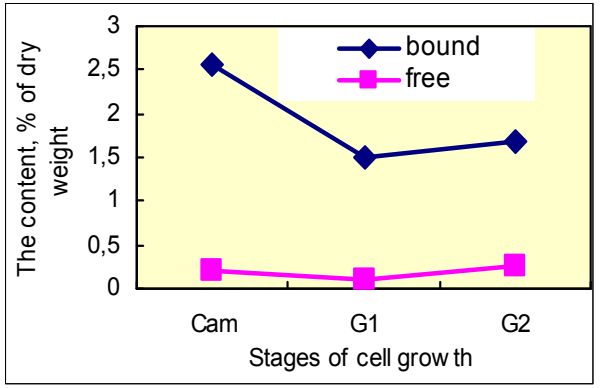

b

Rate of radial growth

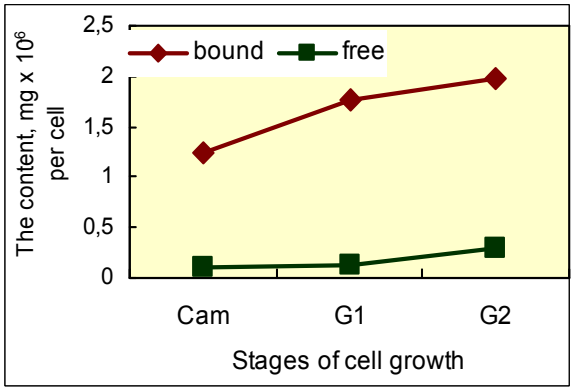



Antonova and Shebeko, Lesovedenie, 1985

c

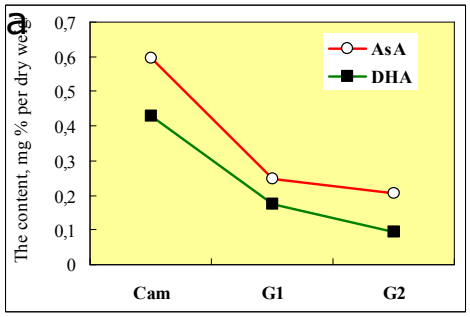



d

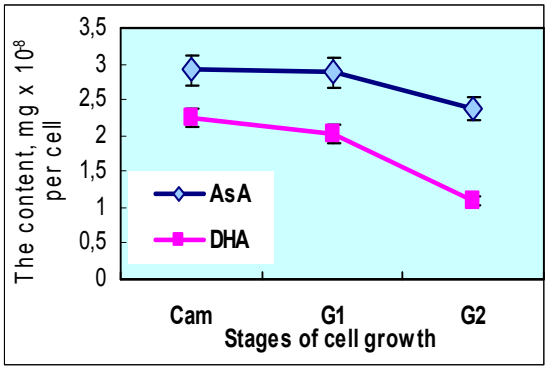


The content of ascorbic (AsA) and dehydroascorbic acids (DHA) (a- per dry weight, b –per cell) and the ratio of AsA/DHA in expansion growth zone

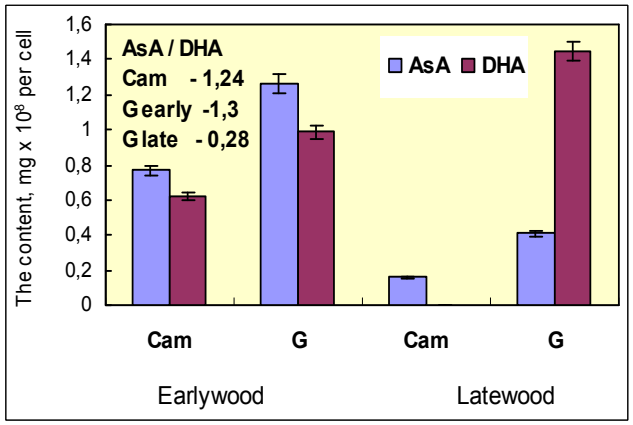
a




b




c




**Radial diameters of tracheids:**

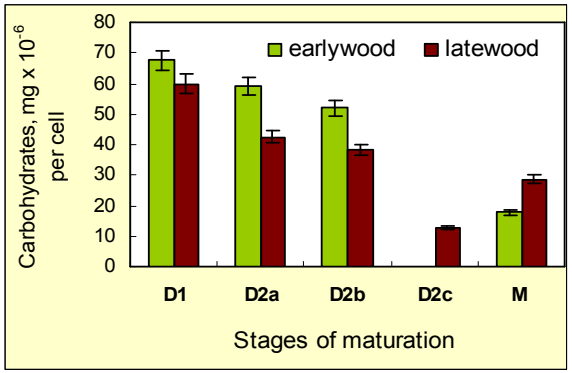
**early xylem - 61.3 mm**

**late xylem - 28.9 mm,**

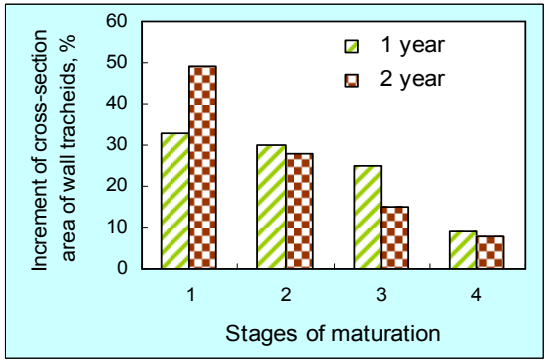
In accordance with the ration of AsA/DHA the radial sizes of latewood tracheids are less than early cells.

Carbohydrates (a - per cell), increment of cell wall cross-section area (b), and of cellulose (c) at consecutive stages of tracheid maturation in larch stem

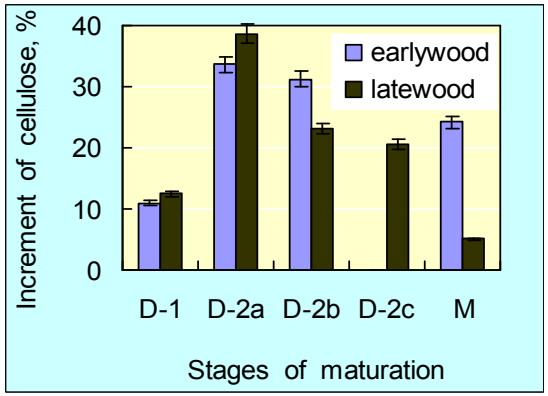
a




b



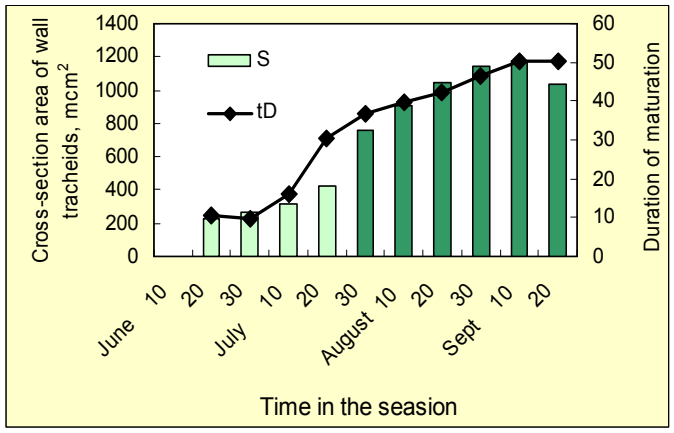

c



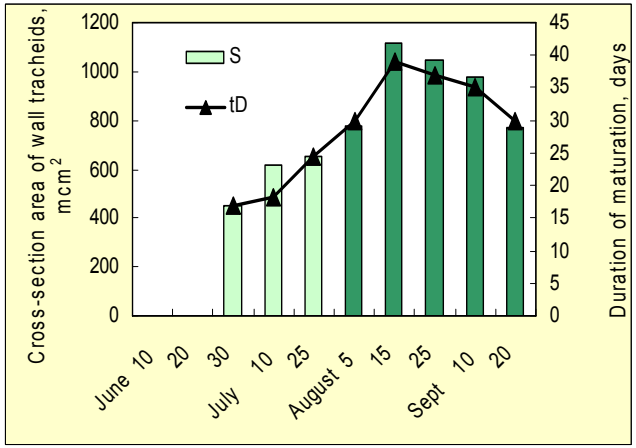

Secondary wall development of earlywood cells doesn't be limited by substratum.

The question is – what influences on wall thickness?

# Cell wall cross-section area (S) and duration of development (tD) of larch tracheids in secondary thickening zone in the course of season



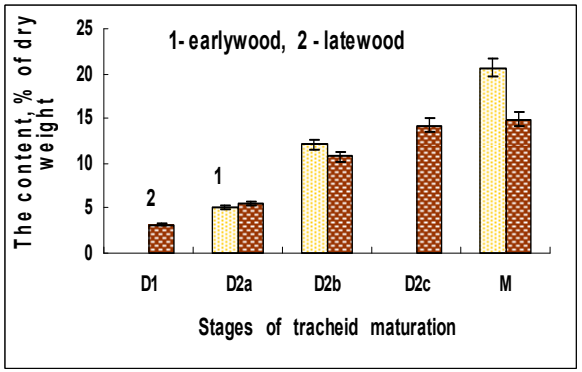

**S** – cross-section area of wall tracheids of **earlywood** **latewood**



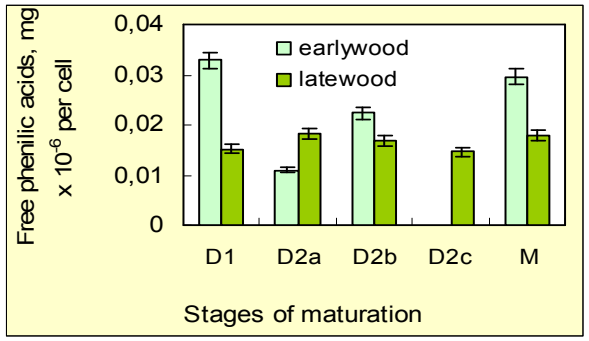

**tD** – duration of development of tracheids in maturation zone:
earlywood - 10 -25 days
latewood - 30 – 50 days

**Cell wall thickness depends on duration of cell development in maturation zone, what in turn depends on a presence of some phenolic acids**

# Deposition of lignin (a), increment of lignin (b), the content of phenolic acids (c) and the ratio of AsA/DHA at consecutive stages of tracheid maturation



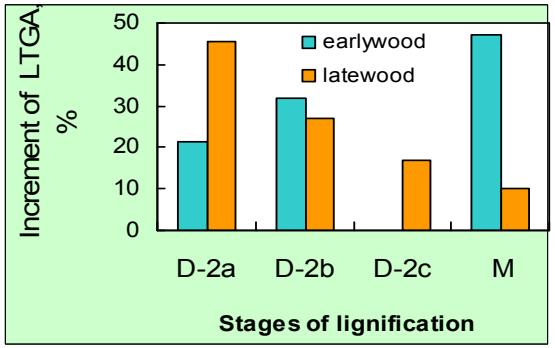

a




c

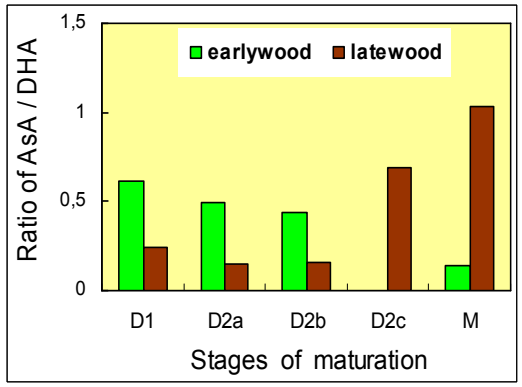
**Free hydroxycinnamic acids**:
**earlywood: p-coumaric ferulic synapic**
**latewood: p-coumaric ferulic synapic**

**Increment of lignin**




b

the ratio of AsA/DHA



# Conclusion

- The formation and development of tracheids of early and late xylem in annual increment occur in different physiological and biochemical conditions what influence their morphological parameters.

- Radial growth of early and late tracheids doesn't be limited by substratum (carbohydrates) and depends in general on the relationship of ascorbic-dehydroascorbic acids what affects on coupling of carbohydrate polymers and consequently on growth of cells.

- The amount and fractional composition of phenolic acids and, in particular, hydroxycinnamic acids, taking part in coupling of polysaccharides in primary walls, and are precursors of lignin, change during developmental stages in agreement with the rate of both growth and lignification of early and late xylem cells.

- The content of AsA and its accessibility to oxidation in cells of the zones of cambium, expansion growth and maturation are resulted in different the number of cells, in different radial diameter, in different wall thickness of early and late tracheids as well as in the rate of lignification of two type of xylem during formation annual ring in larch.