

THE RELATIONSHIP BETWEEN GROWTH-RING WIDTH AND RAY DENSITY AND RAY HEIGHT IN CELL NUMBER IN THE EARLYWOOD OF *PINUS HALEPENSIS* AND *PINUS PINEA*

by

Simcha Lev-Yadun

Department of Natural Resources, Agricultural Research Organization, The Volcani Center,
P. O. Box 6, Bet Dagan, 50250, Israel

SUMMARY

The relationship between growth-ring width and ray size and number was studied in five 24-year-old trees of *Pinus halepensis* Mill. and five 16-year-old trees of *Pinus pinea* L. All trees of both *Pinus* species showed a gradual tendency for an increase in ray height, from an average of less than 4 cells near the pith to 7 or 8 cells in the outer rings. Ray number decreased from more than 70 rays per mm² near the pith to about 40 rays per mm² in the outer rings. No significant correlation was found between growth-ring width and the number of rays per mm² or height of rays for three out of five trees of *P. halepensis* or for any of the five *P. pinea* trees. I conclude that there is no general direct relationship between growth-ring width and ray number and size.

Key words: Cambial activity, differentiation, growth rings, vascular rays, *Pinus halepensis*, *Pinus pinea*.

INTRODUCTION

The ray system is the radial component of the secondary plant body (Philipson et al. 1971). Two types of initials exist in the cambium: the fusiform initials, which divide and differentiate to form the axial system, and the ray initials, from which the radial system differentiates. Fusiform cambial initials can change into ray initials and *vice versa* (Barghoorn 1940a, b, 1941; Philipson et al. 1971; Larson 1994). The ratio of fusiform to ray initials varies greatly among species, as well as within the same plant at different locations and following wounding. These ratios range from 100% fusiform initials in rayless plants, to only 25% fusiform initials in *Dillenia indica* L. (Ghouse & Yunus 1974). In conifers, the average percentage of fusiform initials is about 90%, and in dicotyledons about 73% (Iqbal & Ghouse 1990).

A normal aspect of ontogeny in woody plants is a gradual increase in ray length and width with age, with distance from the pith, and with distance from young leaves (Lev-Yadun & Aloni 1995). This gradual change in ray size and number should be considered when interpreting experimental studies of ray differentiation. Otherwise, ontogenetic changes might be considered the result of the experimental manipulation (Lev-Yadun & Aloni 1991a, 1995).

The influence of the rate of cambial activity on the formation and dimensions of rays has been studied in only a few species, with contrasting results. Bannan (1937) found that in *Thuja occidentalis*, *Taxodium distichum*, *Abies balsamea*, *Tsuga canadensis*, *Larix laricina*, *Pinus strobus*, *Pinus banksiana*, two species of *Araucaria*, three of *Juniperus* and four of *Picea*, the more vigorous specimens had higher rays at the same distance from the pith, while the less vigorous specimens (narrower rings) have more but lower rays. Further research on *Thuja occidentalis* revealed a positive correlation between height, number and total ray volume and ring width (Bannan 1954). Ray height was positively correlated with ring width in *Picea glauca*, *Pinus resinosa* and *Pinus strobus* (Bannan 1965), but the increase in ray height reached its maximum when ring width approached 2–4 mm.

White (1962) found more and wider rays in the faster-growing lower side of a branch of *Sassafras officinale*. The effect of the larger radius on the size (height or width in cell number, or in μm) and number of rays was not considered. In *Fraxinus excelsior* and *Castanea sativa* there were more rays per unit area in wood with wide growth rings than there were in wood with narrower rings. Wider growth rings also had more and wider rays (wider both in cell number and size) in *Fraxinus* and in *Sassafras*, and had an increase in both ray and cell number in *Castanea*. No consistent relationship was found between radial growth rate as indicated by ring width and ray height in these species (White & Robards 1966). Gregory and Romberger (1975) studied the relationships between rate of cambial activity and ray height in *Picea glauca* and *Abies balsamea*. They found a strong positive correlation between annual growth-ring width and number of ray cells per unit area in tangential section, but no analogous correlation with the number of rays. In *Acer saccharum*, an increase in ray size (number of cells per ray), but not in ray number, occurred with increased cambial activity (Gregory 1977); however, the influence of the length-on-age trend on ray size and number was not taken into consideration.

In this work, I studied the relationships between growth-ring width, and the size and number of rays. I show that no general relationships exist between them for young trees of two *Pinus* species.

MATERIALS AND METHODS

Five 24-year-old planted trees of *Pinus halepensis* Mill. and five 16-year-old trees of *Pinus pinea* L. of an unknown genetic background, all growing within several hundred metres of each other, in the western part of the central mountain belt of Israel (about 350 m above sea level), were felled, and discs were taken 20–30 cm above ground level. There are no data available for these two species about when the transition from juvenile to mature wood occurs. The trees grew on Rendzina soil, in a region receiving an average annual precipitation of c. 550 mm. The major factor controlling growth-ring width in these pine species in the region is the annual precipitation (Yakir et al. 1996). To avoid the influence of environmental variability on tree growth, the tree-ring chronologies of the annual growth rings of trees of the two spe-



Fig. 1. Tree ring chronologies of the *Pinus halepensis* (bold line) and *Pinus pinea* (dashed line) trees used for this study.

cies were compared (Fig. 1). Because the trees showed the same growth pattern, it was assumed they responded similarly to environmental factors.

Cross sections were prepared from a narrow sector along one random radius from pith to cambium of each tree, and tangential sections were prepared from the earlywood (near the beginning of each growth ring) of the same sector of each sample. The sections were stained with safranin and fast-green, and mounted in Canada balsam. The width of each growth ring was measured, and a master chronology was prepared for each species.

The number of rays (including fusiform rays) was manually counted in twenty 1.0 mm² microscopic screens in tangential longitudinal sections of each growth ring (110,060 rays in *Pinus halepensis*; 77,600 rays in *Pinus pinea*). Ray height in number of cells was measured by counting the number of cells per ray in a hundred random sequent rays (including fusiform rays) in tangential longitudinal sections of each growth ring (65,770 ray cells in *Pinus halepensis*; 54,295 ray cells in *Pinus pinea*). Because the outer rings of some trees of *P. halepensis* had been wounded, and wounding is known to change ray size and number in *Pinus* (Lev-Yadun & Aloni 1991b, 1992), the counts of these rings were not used for calculations. Therefore, Figures 2 and 4 end with ring 20.

For statistical analysis of single trees, a linear regression (Statistix 1987) was used, and for groups of trees, analysis of variance and covariance with repeated measures (BMDP 1988).

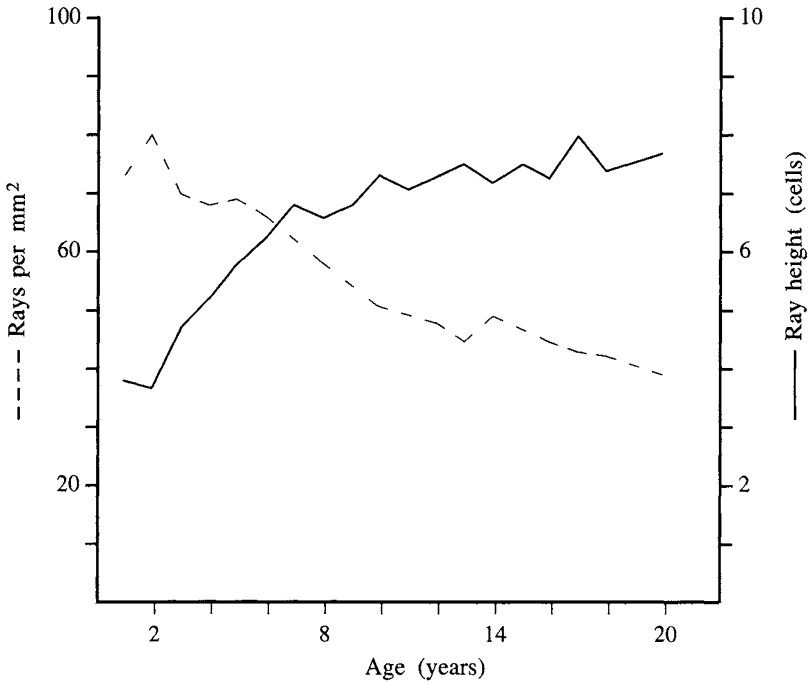


Fig. 2. Average ray characteristics in five trees of *Pinus halepensis*. Dashed line = ray number in mm². Bold line = ray height in cells.

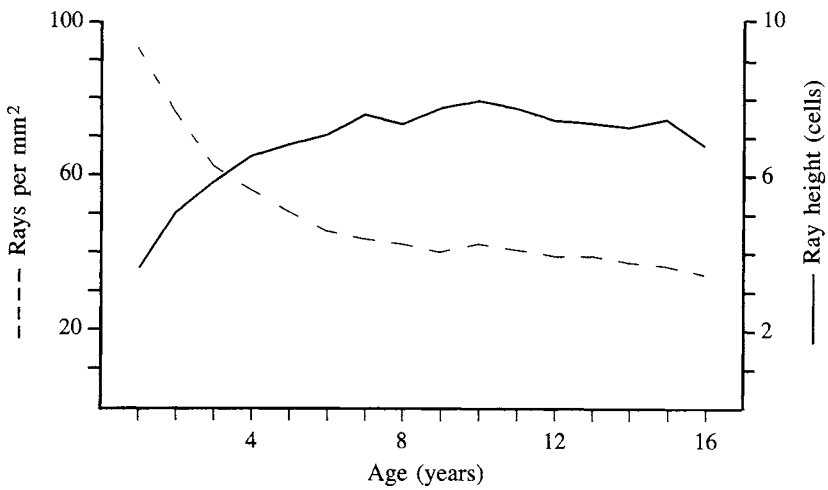


Fig. 3. Average ray characteristics in five trees of *Pinus pinea*. Dashed line = ray number in mm². Bold line = ray height in cells.

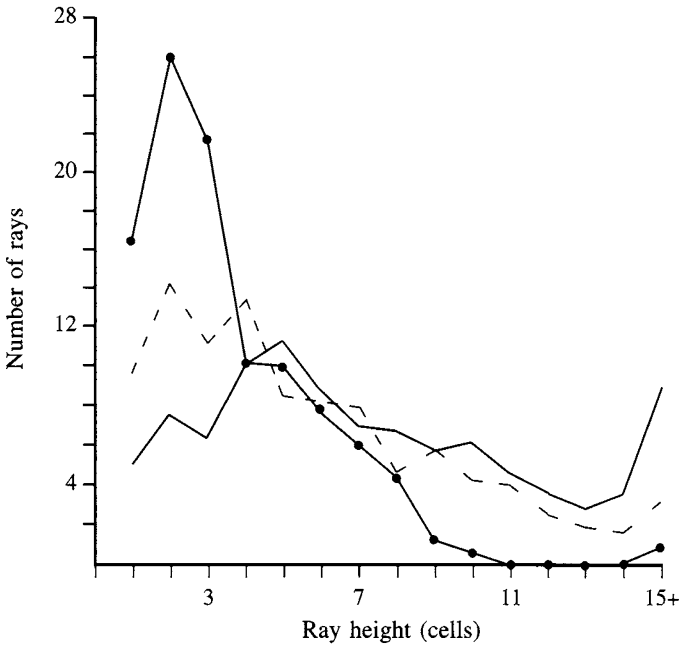


Fig. 4. Distribution of ray heights (measured in cells) in *Pinus halepensis*. Line with bold circles = ring 1. Dashed line = ring 5. Bold line = ring 20.

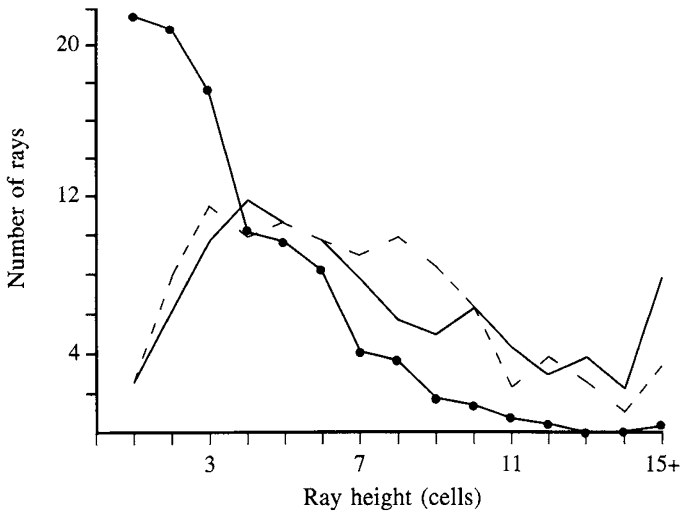


Fig. 5. Distribution of ray heights (measured in cells) in *Pinus pinea*. Line with bold circles = ring 1. Dashed line = ring 5. Bold line = ring 15.

RESULTS

All trees of both *Pinus* species showed a gradual increase in ray height and a decrease in ray number per mm² from the pith outward. The first two inner rings of both species had an average of more than 70 rays per mm², whereas the outer rings had an average of about 40 rays per mm². The average height of the rays (in number of cells) was less than 4 cells in the inner rings, and fluctuated in the outer ones between 7 and 8 cells in *P. halepensis* and around 7 cells in *P. pinea* (Fig. 2 & 3). The frequencies of ray heights also showed a gradual change with age, and more rays were higher than 15 cells when the trees matured (Fig. 4 & 5).

Only for trees 1 and 2 of *P. halepensis* was there a statistically significant positive correlation between growth-ring widths and the number of rays per mm². The correlation was high for tree 1 and low for tree 2 (Table 1). There was a statistically significant positive, but low, correlation between ring width and ray height for trees 1 and 2 of *P. halepensis*. For the other *P. halepensis* trees, and for all *P. pinea* trees, no statistically significant correlation was found (Table 1).

Table 1. Correlation between growth ring widths and ray number and height in *Pinus halepensis* and *Pinus pinea*.

	Tree 1	Tree 2	Tree 3	Tree 4	Tree 5
<i>Pinus halepensis</i> (ray number)					
n	24	23	24	21	22
r ²	0.6515	0.3083	0.0202	0.0164	0.0604
p	0.0000*	0.0060*	0.5081	0.5805	0.2704
<i>Pinus halepensis</i> (ray height)					
n	24	23	24	21	22
r ²	0.4212	0.2433	0.0058	0.0252	0.0081
p	0.0006*	0.0168*	0.7237	0.4919	0.6907
<i>Pinus pinea</i> (ray number)					
n	15	16	16	16	15
r ²	0.0225	0.1167	0.0084	0.0063	0.1656
p	0.5936	0.1953	0.7356	0.7710	0.1322
<i>Pinus pinea</i> (ray height)					
n	15	16	16	16	15
r ²	0.0116	0.0335	0.0000	0.0362	0.1351
p	0.7020	0.4974	0.9804	0.4803	0.1778

n = number of growth rings studied.

* statistically significant at $p < 0.05$.

DISCUSSION

The results from *Pinus halepensis* and *P. pinea* showed that both number of rays in a given area and height of rays were not related to the rate of cambial activity as expressed in growth-ring width. From a total of 191 growth rings from five *P. halepensis* trees and five *P. pinea* trees, ray number and height showed a significant positive correlation with the rate of growth only in the first eight growth rings of two *P. halepensis* trees.

There are contrasting results on the relationships between ray number and the rate of radial growth (= rate of cambial activity). Bannan (1937, 1954, 1965), White (1962), White and Robards (1966), Carmi et al. (1972), and Rao (1988) found a positive correlation between ray number and the rate of cambial activity for some species, while Gregory and Romberger (1975), Gregory (1977), Rao (1988) and Lev-Yadun and Aloni (1991c) did not find such relationships. Kučera and Kučera (1967) found that in *Abies alba* more rays were initiated in summerwood, when the rate of cambial activity was slower. Gregory and Romberger (1975) showed that rays became taller (in cell number) in the latewood of *Abies balsamea*, when growth rate declined.

Several previous reports on the relationships between the rate of cambial activity and ray characteristics (Bannan 1954, 1965; White 1962; White & Robards 1966) gave only partial, or no information on the distance of the narrow and wide growth rings from the pith. Thus, the influence of the length-on-age trend on ray size and frequency (Iqbal & Ghouse 1990; Lev-Yadun & Aloni 1995) cannot be evaluated from those data. The data of Bannan (1937), for branches and roots, are from sections made 3–5 mm outward from the protoxylem, and 100 mm and more for trunks. At such proximity to the pith as studied in branches and roots, every mm or difference in ring age can greatly change the results; a similar problem exists in the trunk data. Moreover, Bannan (1937) concluded that the greater number of rays generally observed in branches as compared with the stem no doubt correlates with the reduced width of the rings and the associated reduction in diameter of the tracheids. The general trend of gradual increase in ray length and width with age, with distance from the pith and young leaves seems to be the outcome of the regulatory role played by gradients of auxin (Lev-Yadun & Aloni 1995). Thus, it may be suggested that Bannan (1937) described a pattern that can be viewed in the light of our understanding of the involvement of phytohormones in regulating cambial activity. The greater number of rays observed in branches as compared with the stem (Bannan 1937) reflects the higher level of auxin flow in young branches as compared with large stems (see Lev-Yadun & Aloni 1993a, b, 1995).

Carmi et al. (1972) were the first to use experimental data rather than developmental observations to examine the relationship between the rate of cambial activity and ray characteristics. The finds of Carmi et al. (1972) for *Ailanthus altissima* triggered further research (Lev-Yadun & Aloni 1991a, b, c, 1992, 1993a, b, 1995; Lev-Yadun 1994a, b), and the progress achieved in these studies concerning the regulatory mechanisms of ray structure helps in evaluating data from previous studies on the relationship between the rate of cambial activity to ray characteristics. Wider growth rings do

not necessarily indicate a higher rate of radial growth since a longer period of cambial activity can also result in wider growth rings. However, when the difference between growth-ring widths are larger than the differences between the duration of cambial activity resulting in narrow or wide growth rings, then changes in growth rate do occur. The inner growth rings of trees (juvenile wood) are usually much wider than the growth rings at maturity (Fritts 1976). However, the rays in the inner growth rings, where the rate of cambial activity is usually higher than in the outer ones, are smaller. Because of all the facts mentioned above, and because the influences of wounding in experiments on ray size and number was not usually taken into consideration, I conclude that the 'black boxes' which are referred to as 'the rate of radial growth' or 'growth-ring width' are not the regulating mechanism of ray size and number.

ACKNOWLEDGEMENTS

I thank the Forestry Division of the Jewish National Fund for supplying the trees. I thank Yigal Avivi, Gideon Grafi, T.F. Shupe and an anonymous reviewer for their comments on the manuscript. Parts of this study were conducted while being at the Department of Botany, The George S. Wise Faculty of Life Sciences, Tel Aviv University and while the author was a Recipient of a Sir Charles Clore Post-Doctoral Fellowship at the Department of Plant Genetics, The Weizmann Institute of Science.

REFERENCES

- Bannan, M.W. 1937. Observations on the distribution of xylem-ray tissue in conifers. *Ann. Bot. n.s.* 1: 717–726.
- Bannan, M.W. 1954. Ring width, tracheid size, and ray volume in stem wood of *Thuja occidentalis* L. *Can. J. Bot.* 32: 466–479.
- Bannan, M.W. 1965. Ray contacts and rate of anticlinal division in fusiform cambial cells of some Pinaceae. *Can. J. Bot.* 43: 487–507.
- Barghoorn, E.S. Jr. 1940a. Origin and development of the uniseriate ray in the Coniferae. *Bull. Torrey Bot. Club* 67: 303–328.
- Barghoorn, E.S. Jr. 1940b. The ontogenetic development and the phylogenetic specialization of rays in the xylem of dicotyledons. I. The primitive ray structure. *Amer. J. Bot.* 27: 918–928.
- Barghoorn, E.S. Jr. 1941. The ontogenetic development and phylogenetic specialization of rays in the xylem of dicotyledons. II. Modification of the multiseriate and uniseriate rays. *Amer. J. Bot.* 28: 373–382.
- BMDP, 1988. BMDP statistical software: pp. 1081–1114. University of California Press, Berkeley.
- Carmi, A., T. Sachs & A. Fahn. 1972. The relation of ray spacing to cambial growth. *New Phytol.* 71: 349–353.
- Fritts, H.C. 1976. *Tree rings and climate*. Academic Press, London.
- Ghouse, A.K.M. & M. Yunus. 1974. The ratio of ray and fusiform initials in some woody species of the Ranalian complex. *Bull. Torrey Bot. Club* 101: 363–366.
- Gregory, R.A. 1977. Cambial activity and ray cell abundance in *Acer saccharum*. *Can. J. Bot.* 55: 2559–2564.
- Gregory, R.A. & J.A. Romberger. 1975. Cambial activity and height of uniseriate vascular rays in conifers. *Bot. Gaz.* 136: 246–253.

- Iqbal, M. & A.K.M. Ghouse. 1990. Cambial concept and organisation. In: M. Iqbal (ed.), *The vascular cambium: 1–36*. Research Studies Press Ltd., Taunton, and John Wiley & Sons Inc., New York.
- Kučera, L. & J. Kučera. 1967. Anatomische Studie über die Entwicklung und Verteilung der Markstrahlen bei der Tanne (*Abies alba* Mill.) I. Charakteristik der Anfangsentwicklung des Markstrahles. *Drevarsky Vyskum* 12: 179–189.
- Larson, P.R. 1994. *The vascular cambium: Development and structure*. Springer-Verlag, Berlin.
- Lev-Yadun, S. 1994a. Experimental evidence for the autonomy of ray differentiation in *Ficus sycomorus* L. *New Phytol.* 126: 499–504.
- Lev-Yadun, S. 1994b. Radial fibres in aggregate rays of *Quercus calliprinos* Webb. – evidence for radial signal flow. *New Phytol.* 128: 45–48.
- Lev-Yadun, S. & R. Aloni. 1991a. Polycentric vascular rays in *Suaeda monoica* and the control of ray initiation and spacing. *Trees, Structure and Function* 5: 22–29.
- Lev-Yadun, S. & R. Aloni. 1991b. An experimental method of inducing ‘hazel’ wood in *Pinus halepensis* Mill. (Pinaceae). *IAWA Bull. n. s.* 12: 445–451.
- Lev-Yadun, S. & R. Aloni. 1991c. Natural and experimentally induced dispersion of aggregate rays in shoots of *Quercus ithaburensis* Decne. and *Q. calliprinos* Webb. *Ann. Bot. n. s.* 68: 85–91.
- Lev-Yadun, S. & R. Aloni. 1992. The role of wounding in the differentiation of vascular rays. *Int. J. Plant Sci.* 153: 348–357.
- Lev-Yadun, S. & R. Aloni. 1993a. Effect of wounding on the relations between vascular rays and vessels in *Melia azedarach* L. *New Phytol.* 124: 339–344.
- Lev-Yadun, S. & R. Aloni. 1993b. Variant secondary growth in old stems of *Ephedra campylopoda* C.A. Mey. *Bot. J. Linn. Soc.* 112: 51–58.
- Lev-Yadun, S. & R. Aloni. 1995. Differentiation of the ray system in woody plants. *Bot. Rev.* 61: 45–84.
- Philipson, W.R., J.M. Ward & B.G. Butterfield. 1971. *The vascular cambium*. Chapman & Hall, London.
- Rao, K.S. 1988. Cambial activity and developmental changes in ray initials of some tropical trees. *Flora* 181: 425–434.
- Statistix, PC DOS 2.0. 1987. NH Analytical Software, Roseville.
- White, D.J.B. 1962. Tension wood in a branch of *Sassafras*. *J. Inst. Wood Sci.* 2: 74–80.
- White, D.J.B. & A.W. Robards. 1966. Some effects of radial growth rate upon the rays of certain ring-porous hardwoods. *J. Inst. Wood Sci.* 17: 45–52.
- Yakir, D., S. Lev-Yadun & A. Zangvil. 1996. El Niño and tree growth near Jerusalem over the last 20 years. *Global Change Biology* 2: 97–101.