



Cone and Seed Improvement Program BCMoF Tree Seed Centre

from Tree Seed Working Group Newsbulletin
#42 2005



Resin Vesicles in Conifer Seeds

The term resin vesicles (or resin cavities) is commonly understood in conifer seed technology, but it is amazing how little is known about these structures. This is surprising considering the influence these structures can have on successful germination. This article attempts to summarize the known literature of resin vesicles in conifers. I would appreciate knowing of any additional documentation.

Resin vesicles are found within seeds of the following genera in the Pinaceae: *Abies*, *Cedrus*, *Keteleeria*, *Nothotsuga*, *Pseudolarix* and *Tsuga* (Frankis 1988) and within the genera *Libocedrus* (Schopmeyer 1974) and *Thuja* in the Cupressaceae. Within the Pinaceae those genera bearing resin vesicles diverged from those not bearing resin vesicles quite early in evolutionary history (Wang et al. 2000). Singh (1978) discusses the “differentiation of mucilage canals (cycads) or resin ducts (in conifers)” in the section on development of the seed coat providing a hint to the origin of these structures. The occurrence of resin vesicles in *Thuja* is not well documented!

The resin vesicles in *Abies* spp. appear to differentiate at the young archegonial stages in *Abies grandis* (Singh and Owens 1982); during the last stages of female gametophyte development in *Abies amabilis* (Owens and Molder 1977); and soon after fertilization in *Abies lasiocarpa* (Singh and Owens 1981). The vesicles consist of an oblong cavity surrounded by a layer of epithelial cells described as richly protoplasmic. The enlargement of the resin cavities causes depressions on the surface of the female gametophyte (Singh and Owens 1982; personal observation). This depression causes a displacement of nutritive tissue and although the role of resin vesicles is not understood – this tradeoff in potential energy indicates it must have an important role in the success of the species that possess them.

The resin arising from other parts of the tree have been studied more fully than resin from seeds. This trend will probably continue as more information is gained on the role of terpenoids from resin and how to induce traumatic resin defenses against herbivores and pathogens (i.e. Faldt et al 2003). Hopefully this will also increase our knowledge on resin vesicles and their role in conifer seed. For those really looking for more on resin a new book entitled “Plant Resins: Chemistry, Ecology and Ethnobotany” by Jean H. Langenheim was published in 2003. Reference to a review of this book by Gershenzon (2003) is included for those wishing to explore the subject further.

Resin is considered “a lipid-soluble mixture of volatile and non-volatile terpenes and/or secondary compounds that are secreted in specialized structures”(Langenheim 2003). The amount of resin in *Abies alba* has been documented as up to about 20% of the fresh mass of seeds with monoterpenes accounting for 90% of the total volume of volatile terpenes. The monoterpenes have been studied and the main components and their [ranges] are: limonene [60 - 90%]; α -pinene [5-30%]; Δ -3-carene [3-4%] and β -pinene [1-2%] (Čermák 1987). During storage for 24 months at 1-3° C the monoterpene composition changed with α -pinene decreasing and Δ -3-carene decreasing (Čermák and Penka 1978).

The most detailed studies were done by Čermák (1987) who looked at variability in monoterpenes between and within trees and in relation to the position of the resin vesicles on the seed surface. Differences between trees were significant, but cones from the same tree differed only slightly. There was actually greater variability found between different resin vesicles on the same seed compared to different cones (if the same resin vesicles are compared). These results indicate the importance of clearly defining the location of resin vesicles in further studies. Čermák (1987) determined the resin vesicles closest (abaxial) and furthest (adaxial) from the ovuliferous scale had quite different proportions of α -pinene, β -pinene and limonene forming a gradient around the seed. No differences between resin vesicles were noted for Δ -3-carene.

The reduced germination from seedlots with damaged resin vesicles is well-known and has been duplicated through puncturing resin vesicles or dipping the seed in the resin (Gunia and Simak 1968; Kitzmiller et al 1973; Kitzmiller et al 1975). The resin has also been shown to be inhibitory to germination of pine and spruce (Rohmeder 1951). It is especially interesting that if resin vesicles were damaged following stratification (*vs.* before) that the decrease in germination was not as great (Gunia and Simak 1968; Kitzmiller et al 1973) and that damaged seed performed better than undamaged seed without prechilling (Kitzmiller et al 1975). Interesting findings on a few seedlots and surprising that no follow-up studies have been done in this area.

A question that many BC nurseries are asking is whether there is good seed sanitation method for *Abies* spp. Some early work with hydrogen peroxide indicated that the proportion of fungi on the seed coat could be reduced with hydrogen peroxide, but in general (one exception) a decline in germination was found (Edwards and Sutherland 1979). A trial involving seedlots [# in brackets] of *Abies amabilis* [6], *A. lasiocarpa* [2] and *Abies grandis* [2] were exposed to 3% hydrogen peroxide for 0.5, 2 and 4 hours after soaking and to a 2-hour treatment performed prior to soaking. Data was collected on resin vesicle damage, germination capacity and incidence of *Fusarium* sp. and *Caloscypha fulgens*. The results overwhelmingly showed the individual seedlot accounted for the greatest proportion of variation in germination capacity [83%]; resin vesicle damage [78%]; *Fusarium* contamination [85%] and *Caloscypha fulgens* infection [86%]. In *Amabilis* fir a significant increase in resin vesicle damage was found by extending the sanitation treatment from 0.5 to four hours with a decrease in germination from 72 to 66% (not statistically significant) (Kolotelo 2001). Growers are currently cautioned against the use of hydrogen peroxide on resin vesicle species due to the uncertainty of the practice. Some growers have successfully used hydrogen peroxide to sanitize *Abies* spp., but others have had very poor results from the practice. The use of running water soaks to reduce pathogen loads is considered the best alternative at this time. This can help with *Fusarium* which is borne on the seed coat, but it would have no impact on *Caloscypha fulgens* which infects and kills the seed. There is still lots of work that needs to be done on this issue, but work on *Abies* spp. in general does not rate very high in many jurisdictions today.

Several theories have been proposed for the role of resin vesicles. Based on the fact that damaged seed without a prechill germinates better than undamaged seed without a prechill, it was hypothesized that seedcoat dormancy is an important factor and that resin vesicle damage was possibly a form of scarification (Kitzmiller et al 1975). There may be a relationship, but undamaged and stratified seed obtained the highest germination in all studies referred to here. It was also suggested that the resin vesicles were involved in dormancy and prevented germination in the same year as seed dispersal (Rohmeder 1951) and that the resin provides a layer of

protection preventing excessive drying of the embryo and megagametophyte (Gunia and Simak 1968). Still lots of mystery regarding resin vesicles and their roles in conifer seeds – hopefully this has brought some of the literature to people's attention and maybe someone would be interested in looking at some of these unanswered or unasked questions. I have connections for seed if anyone is interested ☺

References

Čermák, J. 1987. monoterpene hydrocarbon contents of the resin from seeds of silver fir (*Abies alba* Mill.). *trees* 1:94-101.

Čermák, J. and Penka, M. 1978. quantitative variation of monoterpene hydrocarbon composition in resin from *Abies alba* Mill. Seeds during their longterm storage. *Biologia (Bratislava)* 33:565-572.

Edwards, D.G.W. and J.R. Sutherland. 1979. Hydrogen peroxide treatment of *Abies* seeds. Environment Canada forestry service Bi-monthly research Notes 35910:3-4.

Faldt, J., D. Martin, B. Miller, S. Rawat, and J. Bohlmann. Traumatic resin defenses in Norway spruce (*Picea abies*): methyl jasmonate-induced terpene synthase gene expression, and cDNA cloning and functional characterization of (+)-3-carene synthesis. *Plant Mol. Biol.* 51:119-133.

Frankis, M.P. 1988. Generic inter-relationships in Pinaceae. *Notes Royal Botanical Garden Edinburgh* 45 (3): 527-548.

Gershenzon, J. 2003. Book Review. *Plant Resins: Chemistry, Ecology and Ethnobotany* by Jean H. Langenheim. Timber Press. *Journal of Chemical Ecology* 29:2777-2779.

Gunia, S. and M. Simak. 1970. Effect of damaging resin vesicles in the seed coat on the germination of silver fir (*Abies alba* Mill.) seeds. In *Proc. International Symposium on seed physiology of Woody Plants*. Kornik, Poland Swept. 3-8, 1968. pp 79-83.

Kitzmiller, J.H., J.M. Battigan and J.A. Helms. 1973. Effect of resin vesicle damage on germination of stored *Abies concolor* seed. True Fir Management Co-Operative. Internal Report #1. 16 pp.

Kitzmiller, J.H., J.A. Helms and P. Stover. 1975. Influence of seed treatment on germination of *Abies concolor* and *A. magnifica*. True Fir Management Co-Operative. Internal Report #3. 39 pp.

Kolotelo, D. 2001. Seed sanitation methodology for *Amabilis* fir. Forest Genetics Council of BC Tree Improvement Program project Report 2001/2002. pp 63-64.

Langenheim, J.H. *Plant Resins: Chemistry, Ecology and Ethnobotany*. Timber Press, Portland, OR ISBN 0-88192-574-8.

Owens, J.N. and M. Molder. 1977. Sexual reproduction of *Abies amabilis*. *Can. J. Bot.* 55:2653-2667.

Rohmeder, E. 1951. Beitrage zur Keimungsphysiologie der Forstpflanzen. Bayer. Landwirtschaftsverlag, Munchen {quoted from Gunia & Simak1968}

Schopmeyer, C.S Tech Co-ord. 1974. seeds of Wood Plants in the United States. Agricultural handbook No. 450. Forest Service USDA. Washington, DC. 883 pp.

Singh, H. 1978. Embryology of gymnosperms. Gebrüder Borntraeger, Berlin, Stuttgart

Singh, H. and J.N. Owens. 1981. Sexual reproduction in subalpine fir (*Abies lasiocarpa*). Can. J. Bot. 59:2650-2666.

Singh, H. and J.N. Owens. 1982. sexual reproduction in grand fir (*Abies grandis*). Can. J. Bot. 60:2197-2214.

Wang, X-Q, D.C. Tank, and T. Sang. 2000. Phylogeny and divergence times in Pinaceae: Evidence from three genomes. Mol. Biol. Evol. 17(50:773-781.

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