Septoria spp. Infection Among North American Northwest Coast Provenances of Black Cottonwood (*Populus trichocarpa*): A Field Study of Natural Infection in a Fraser Valley Plantation

April 11th, 2011

Tomas Cimolai

FRST 499 – B.S.F. Thesis

Abstract

Given the recent finding of *Septoria musiva* among poplars in a Fraser Valley plantation and nearby vicinity in British Columbia, we undertook a field study of *Septoria* spp. presence among black cottonwood (*P. trichocarpa*) trees which were being assessed in a British Columbian provenance study. For seven such provenances, *S. musiva* and *S. populicola* were found on leaves in frequencies of 3.9-33.3% and 65.8-85.3% respectively. There were no significant differences in the isolation frequency between provenances for either fungus ($p \geq 0.10$). The provenances were then pooled into two large groups to account for higher and lower latitudes; the lower latitude (South Coast/USA) cluster was found to bear both *S. musiva* and *S. populicola* in lesser frequencies ($p=0.03$ and $p=0.01$ respectively). *S. musiva* was isolated from tree canker, but the presence of canker-like lesions on stem and branches did not correlate with the presence of either fungus overall ($p>0.10$). Endemic *S. musiva* among poplars in this plantation raises several concerns about the local epidemiology. Further studies to confirm our findings are warranted as are studies that further the understanding of the *S. musiva-P. trichocarpa* pathosystem.

Key words: *Septoria musiva*, *Septoria populicola*, *Populus trichocarpa*, British Columbia

Introduction

The black cottonwood poplar (*Populus trichocarpa*) is among the dominant trees in coastal British Columbia, although its presence extends beyond the latter region in this province. Known as the 'giant cottonwood', it has tremendous growth potential both for natural and plantation environments. The native species and its hybrids are of value economically in several wood and pulp products. It is of great significance to future carbon sink endeavours. The biology of this tree is detailed in the accompanying review (Cimolai, 2011).

Like other poplars, *P. trichocarpa* is susceptible to many diseases. Among the infectious category of tree diseases, infection of leaves, branches, and stems by *Septoria* spp. has been recognized for decades (Callan, 1998). Leaf spot due to *Septoria populicola* has been acknowledged in the Pacific Northwest as long as *Septoria* spp. disease has been studied (Thompson, 1941; Newcombe et al., 1995). *Septoria musiva* is a well known pathogen causing leaf spot and canker among poplars in the eastern and central regions of North America (Bier, 1939; Waterman, 1946; Waterman, 1954b; Zalasky, 1978; Cellerino,1999; Jones and Benson, 2001). Canker in particular can be quite damaging and has the potential to reduce biomass availability (Cimolai, 2011). In commercial plantations, *S. musiva* canker can be especially devastating. In British Columbia, it has been generally believed that *S. musiva* is not a prevalent pathogen even though it has been experimentally shown that the fungus can infect and canker *P. trichocarpa* (Bier, 1939; Bier, 1961; Newcombe et al. 1995).

The potential role for *S. musiva* in black cottonwood disease has more recently been raised to a higher level of alert in British Columbia with the advent of two key reports. Callan et al. (2007) provided convincing evidence for the presence of *S. musiva* in Fraser Valley plantation hybrids through both culture and molecular studies in 2006. A review of the citations of *S. musiva* as noted in the herbarium catalogue found at the Pacific Forestry Centre (see web-site below in reference list), however, denotes samples which may have borne *S. musiva* as early as 1988-1991. Of note, among the latter samples, the sources have included the same Fraser Valley area which was investigated by Callan et al. (2007). Beauseigle et al. (2010) have now added to the latter findings by determining the presence of *S. musiva* among the same Fraser Valley area black

cottonwoods and hybrids, but noting that limited spread may also have occurred among native black cottonwoods in the same general region. During the latter work, *S. musiva* was found among trees from a *P. trichocarpa* provenance trial.

The existing provenance trial as indicated above provides the basis for several potential studies of the *S. musiva-P. trichocarpa* pathosystem. In this study, we made use of the latter resource by conducting a field survey for *Septoria* spp. in leaf spots as determined by molecular techniques. With the knowledge that *S. musiva* existed among trees of this black cottonwood provenance trial, we sought to determine a point prevalence of *Septoria* spp. among spotted leaves during an autumn period. This was a pilot study for the purposes of hypothesis generation rather than hypothesis testing.

Methods

Location and Environment – This study was conducted among a black cottonwood plantation in the Fraser Valley within the locale of Harrison Mills, British Columbia (Figures 1 and 2). The plantation has been established for the purposes of various biological studies among British Columbian provenances. The geographic area can be localized as 49.23^{o} latitude and -121.91^{o} longitude. The average rainfall and seasonal temperatures for this region are detailed in Table 1. This site and the vicinity in general has long been an integral part of plantations for commercial purposes and for the development of hybrid poplars. This provenance trial setting is adjacent to the Fraser River but separated by a high dyke. The site is level, has rich soil, and has reasonably good drainage. The water table is necessarily high given proximity to the river. Such a site is consistent with the description of 'riparian'.

Plantation Characteristics – In addition to black cottonwood trees, the underbrush is mainly composed of grasses, and there is admixture with blackberry vines. Such adjacent growth and soil quality is consistent with what Smith (1957) would describe as modest to good quality. The plantation is bordered by small natural access roads, a railway, and the river. The plantation plot is longitudinal mainly and houses well over 1000 trees, some of

which have already succumbed to natural conditions of various sorts. One region of this plantation has reportedly suffered flooding during the early growth years of these trees.

Sources of Black Cottonwood Provenances – *P. trichocarpa* samples were obtained from areas mainly in British Columbia, but also from some sites in Washington State and Oregon due south (CY Xie, British Columbia Ministry of Forests and Range, personal communication). A total of 516 clones were obtained from 35 drainage regions. Figure 3 details the sources of these trees for this particular plantation. For practical purposes, the provenances have been grouped in this study by region, and they are collectively divided into the following seven areas: South Coast Region, Mid-Coast Region, Smithers Region, Dease Lake Region, Far North Region, Prince George Region, and USA Region. A clustering effect of sampling from these regions can be visualized from a review of the map in Figure 3.

The plantation was established in 2007 with one year old rooted cuttings which were raised in a greenhouse environment at the Kalamalka Forestry Centre in Vernon, British Columbia. This plantation has been mainly established for the purposes of exploring and maintaining a catalogue of *P. trichocarpa* genetics.

Characteristics and Plantation Distribution of Provenances – After transport to the Harrison Mills location, trees were distributed by a randomized complete block design with single tree plots in which the initial spacing was approximately 2.5 x 2.5 metres. Depending on the availability of material, each clone was repeated from 4-8 times. No fertilizer was used during or after the planting processes.

Leaf Sampling – Leaf samples were collected from 1070 trees during the fall of 2010 over the period of September 15-30. Each tree was tagged in planting sequence, and the provenance source could be traced to each tag. Individual trees were examined for leaf spot and canker or canker-like lesions on stems and branches. For leaf spot, lesions were sought which possessed characteristics typical of *Septoria* leaf spots (Thompson, 1941; Waterman, 1946; Cooper and Filer, 1976; Zalasky 1978; Palmer et al. 1980; Ostry and McNabb, 1985; Ostry et al. 1989; Callan BE, 1998; Cellerino,1999; Sinclair and Lyon

2005; Beauseigle et al., 2010). Examples of such appearances are detailed in the accompanying review (Cimolai, 2011). Three leaves containing characteristic *Septoria* spots were harvested from the tree and stored in paper lunch bags for transportation to the laboratory. For those trees where typical spots were not evident, leaves were collected which had some form of affliction bearing similar traits. Only a fraction of the trees (n = 297; 27.8%) sampled were thereafter included in this study. This subsampling was proposed by the British Columbia Ministry of Forests, Lands, and Natural Resource Operations.

Canker-Like Lesion Identification – Tree stems and branches were inspected for lesions which could be considered as cankers by the observer (TC). A database of trees having some form of these lesions was maintained for later correlation. Although some branch cankers were sampled, the assessment of such lesions for *Septoria* spp. by culture or genetic detection was not intended to be a routine part of this study.

Laboratory Identification of *Septoria* Infestation – Among the three leaves selected per tree, one of the leaves was chosen, and an isolated leaf spot was subsequently used for detection of infection. The choice of sample was determined by an examination of the available leaves, and the most likely representative leaf spot area was chosen for excision by scalpel. DNA from the spot was extracted as detailed by Beauseigle et al. (2010) with the use of a modified protocol and the QIAGEN DNeasy MiniPlant Kit. Samples were thereafter subjected to a nested polymerase chain reaction (PCR) amplification as detailed previously (White et al., 1990; Feau et al., 2005; Beauseigle et al., 2010). The initial round of amplification produces a >550 base-pair product which corresponds to nuclear rDNA internal transcribed spacer region; universal fungal primers (ITS1F and ITS4) are employed for this step. In the second step of amplification, another universal primer (ITS1) and a *Septoria* specific primer (Septop) are used. After the final amplification, the DNA products are compared to reference sequences for *S. musiva* and *S. populicola*. Confirmation of the identification was achieved with assistance of gene sequencing software. Polymorphisms among the PCR products distinguish species. Culture of the fungi was not conducted in this experiment routinely.

Statistical Analysis – Frequencies of positive samples over the total number of samples assessed were calculated. These frequencies were compared between black cottonwood provenances. The data were also assessed to determine if there was an association of fungal isolation with the appearance of canker-like lesions. Statistical analyses using the g test, chi-square test, and Fisher's exact test were made to assess differences among the study groups. Correction for multiple testing was made for comparisons among data from Table 3 (Bonferroni correction).

Results

Geoclimatics - Table 1 compares location and general weather patterns for various sites in British Columbia. The plantation in this study must be viewed as one that has a temperate climate. Nearby Chilliwack (Figures 1 and 2) is rated as among the most temperate cities in Canada let alone British Columbia. Temperatures can be reasonably high in the mid-summer as is typical for the upper Fraser Valley agricultural region and indeed much higher than many other British Columbia coastal regions. There is a rarity of extreme cold in the winter, consistent with the buffering effect of being relatively coastal. Precipitation is high, and Table 1 illustrates that it has rainfall only second to Prince Rupert among the venues detailed. Both the conditions of temperate climate with warm summers and the high precipitation should favour most fungal pathogens; both of these conditions would typically favour good growth of fungi under laboratory conditions.

Table 2 outlines some rainfall and temperature data that are relevant to the five months preceding this provenance trial and to the five months preceding the acquisition of samples as detailed by Beauseigle et al. (2010). In addition, weather data are shown for the same months but relating to the year so recorded when *S. populicola* was epidemic (Newcombe et al., 1995).

Laboratory Identification – Consistent species polymorphisms were identified by the laboratory method in all positive identification except for three samples in which both

species polymorphisms were determined. *S. musiva* and *S. populicola* were found among leaves from 19/297 (6.4%) and 210/297 (70.7%) trees respectively (Table 3).

Frequencies of *Septoria* spp. Identification – Characteristic leaf spots for *Septoria* spp. were apparent on a large number of trees. Given the fall season when the leaves were sampled for this study, mold infection is not unexpected. The amount of spotting per tree was however quite variable. Figures 4, 5, 6, and 8 exemplify some leaf spotting.

S. musiva isolations varied among the provenances in frequencies from 3.9-33.3% (Table 3). The two higher frequencies among the latter, however, occurred for provenances with small sample sizes. There was no significant difference between these frequencies for provenance to provenance comparisons ($p \geq 0.10$). *S. populicola* isolations varied from 65.8-85.3% (Table 3). Although it is apparent that a higher frequency of *S. populicola* isolation was made from Mid-Coast and Smithers provenances, and although there is a trend towards a significant difference when compared to some other provenances (e.g., South Coast), statistical significance for provenance to provenance comparisons was not achieved when corrected for multiple comparisons ($p > 0.10$).

Given the small sample sizes for some groups, which in themselves leads to a precarious situation for statistical analysis for differences among provenances, a comparison was assessed with the pooling into two clusters. Specifically, the South Coast and USA provenances were grouped, and the remainders were then added together for the second group. These groupings were made to take into account a stratification for latitude and more moderate coastal effects of tree origin. A typical cycle of *P. trichocarpa* phenology is illustrated from trees in Oregon, and it is anticipated that this would be generally similar to that in British Columbia (Boes and Strauss, 1994). In such analyses, both *S. musiva* and *S. populicola* were isolated in lesser frequencies from the South Coast-USA cluster ($p=0.03$ and $p=0.01$; Tables 4 and 5).

Table 2 details the frequencies of *S. musiva* detection for this study in comparison to previous assessments. Individual and average temperatures and precipitation are also detailed for time periods prior to leaf acquisition for the given year. For the two years (2009 and 2010) for which there is *S. musiva* data, the year with the higher temperatures (2009) had greater isolation.

Table 2 also details the frequencies of *S. populicola* detection for this study in comparison to previous assessments. It is evident that the isolation rate from this study exceeded that from other local studies as detailed for 2008 and 2009. There is no clear correlation with temperature or rainfall to account for the latter. As a comparator, climate variables were also obtained for 1993 during which time epidemic *S. populicola* was said to prevail in the Pacific Northwest (Newcombe et al., 1995). It appears that the epidemic 1993 had both warmer temperatures and greater rainfall during April to June when leaf evolution is at its greatest.

Canker Identification and Correlation - Figures 4-8 demonstrate the types of cankers or canker-like lesions so identified. Some of these were sampled for culture, and *S. musiva* was identified. A systematic identification of canker causes was not performed in this study, but it is conceivable that cankers may also be caused by the consequences of abiotic damage or other microbial causes (Callan, 1998). Some of the cankers were associated with distal stem kill and cracking. Both stem and branch cankers were seen. It is not possible to be sure that there cankers in general were caused by some form of mold.

Nevertheless, an analysis was made of fungal identification from trees with and without canker-like lesions (both stem and branch). There was no significant difference in the frequency of these lesions whether *S. musiva* or *S. populicola* was found in the leaves ($p>0.10$; Tables 6 and 7). In another analysis, the frequency of the fungi was examined for trees with canker-like lesions to determine if the lesion location (stem or branch) was associated with either mold. There was no significant difference in the frequency of these lesions whether *S. musiva* or *S. populicola* was found in the leaves ($p>0.10$; Tables 8 and 9).

Discussion

Septoria spp. leaf spot is common among trees in British Columbia and the Pacific Northwest, but such fungal disease has been largely attributed to *S. populicola*. The high frequency of *S. musiva* in this Fraser Valley plantation raises several concerns both in regard to the source and chronology of origin. Based on historical anecdotes from

many in the field, black cottonwood canker due to *S. musiva* had not been cited previously (Bier, 1961; Newcombe et al., 1995). Leaf spot in the Pacific Northwest had been attributed to *S. populicola* only, and our study certainly confirms the high prevalence of this fungus in black cottonwood. Had *S. musiva* been very prevalent, even in occasional epidemic form, one may have expected that some canker in the wild might be identified. As noted in the herbarium catalogue from the Pacific Forestry Centre, samples which may have borne *S. musiva* as early as 1988-1991 are recognized. Callan et al. (2007) then provided more exact science by finding *S. musiva* in Fraser Valley hybrid plantations in the same general area as located in our field trial and as sampled recently by Beauseigle et al. (2010). Beauseigle et al. (2010) found the fungus among natural black cottonwood as well as in plantation stands. The infrequency of *S. musiva* in our field trial does not detract from the fact that the fungus exists in this black cottonwood plantation. There are two ways to reconcile these findings. As *S. musiva* was known to exist among poplars, especially in the central and eastern North America, the fungus may have been imported from other plantations as poplars and their hybrids were received for sampling (Bier, 1942; Strobl and Fraser, 1989; Royle and Ostry, 1995; Sinclair and Lyon, 2005). Once transported from such areas, whether the conditions in British Columbia are favourable for fungal survival or not, the fungus established itself locally. If that were to be the case, importation could have arisen decades ago since poplar silviculture in this area dates back a considerable time. Thereafter, the fungus could have survived and propagated to incoming seedlings or cuttings. The current provenance trial plantation was established in 2007 with one year-old rooted cuttings that were raised in a greenhouse environment at the Kalamalka Forestry Centre in Vernon, British Columbia. The latter might also have been a source for *S. musiva*, and a study of the latter might prove of value to further the understanding of origin. The second, but perhaps less credible, possibility is that *S. musiva* has been in existence in this region, maybe even the entire of British Columbia, for longer than the aforementioned studies would allow us to believe. If, for whatever reason, *S. musiva* is not highly favoured by the local environment, it might yet be capable of maintaining itself in low numbers. If the fungus existed in such low numbers as determined by our field trial, it could conceivably escape the threshold for detection, since the vast majority of leaf spots would be attributed to *S. populicola*. If

S. musiva was rarely causing leaf spot in the wild, it might also be even more rarely causing canker in the wild so that the likelihood of finding *S. musiva*-related canker would be very rare. The lack of a large number of archived leaf spot samples precludes the opportunity to determine whether new molecular techniques could solve this genesis issue. Future studies of native poplar in this province with the use of molecular techniques could shed light on this important ontological aspect.

Importation of infected clones is therefore of great concern. Likewise, it is possible that export of North American, and British Columbian, biologicals could lead to infection in other parts of the world (Riker, 1961). Methods to decontaminate potentially infected cuttings could be of value (Waterman and Aldrich, 1952; Waterman, 1954a; Waterman, 1954b). Surface sterilization of poplar cuttings has some benefit in reducing the fungal load (e.g., mercuric chloride with and without alcohol), but it is nearly impossible to totally remove fungal contamination without seriously injuring the cutting. Therefore, exchange of trees should be approached cautiously.

There were no significant differences for the detection of either fungus from the seven provenances. One must be cautious however in the potential for overinterpretation of these data. Most critical to the limitations inherent in this survey was the small numbers of trees in several of the provenance groupings. Firstly, very small numbers complicate the choice of probability testing for assessing significant differences. Secondly, small changes in the numbers have the potential to significantly change the observed outcomes. Greater numbers of trees per provenance are obviously desirable to ensure a more robust analysis.

The greater apparent frequency of both fungi from provenances of higher latitude must also be viewed cautiously. Although the latter differences may yet prove to be true in future similar research, the actual differences may not truly have relevance to field work. That is, a difference in a few percentages points for isolation rate, even if statistically significant, may not influence much how these findings will be applied for day to day choices of trees for this plantation. Again, future studies to confirm these preliminary findings are warranted. When previously investigating provenances from Oregon, Washington, and British Columbia, the ten best biomass producing clones originated from six different provenances, thus indicating the wide variability of *P.*

trichocarpa in nature (Heilman and Stettler, 1985a). The latter study did not indicate that beneficial biomass production was associated with differences in latitude of origin. Northern provenances have better photosynthetic capability, although they are more photoperiod sensitive (Howe et al., 1995). Photosynthesis is better among trees from lower xeric environments compared to upper mesic environments of the same valley, but generalizing, mesic clones have better photosynthesis than xeric clones (Dunlap et al., 1993). British Columbian provenances have been examined in some detail (Gornall and Guy, 2007). Carbon dioxide assimilation increases with increasing latitude of the provenance but not among clones within that provenance. Stomatal density also correlated with latitude. Northern provenances have increased carbon dioxide assimilation and stomatal conductance to allow for the shorter growing season. The longevity of leaves is inversely related to latitude. As one moves from provenances in the southwest of Oregon and Washington to the northeast towards Washington-British Columbia, it is apparent that the southwest trees have smaller leaves and more numerous branches along with later fall growth (Weber et al., 1985). When comparing black cottonwood from southeastern British Columbia to that from Idaho, it was determined that leaf morphology was not a useful predictor for tree height (Riemenschneider et al., 1994). From studies of the latter, it was proposed that selection of favourable tree characteristics needed to be considered on an individual population basis. Nevertheless, there is plenty of variation even within the same region. For example, leaf size and other measures can be adapted even at a single riparian level along a single slope of forest (Dunlap et al., 1995). Looking at enzyme variation among black cottonwood from the Pacific Northwest, Weber and Stettler (1981) found an overall high genetic similarity among trees from different riparian populations, although there was still considerable differences among clones from the same region. The regions with the most differences were those of large river drainages. Thus, it is apparent that *P. trichocarpa* has responded to climatic selection pressures at both local and regional levels. The association of any of the above with tree susceptibility to *Septoria* spp. is essentially unknown. Nevertheless, such variation based on the aforementioned studies invites multivariate analyses to determine susceptibility.

Over the growing season, there is an apparent upwards migration of *S. populicola* to involve leaves throughout the tree (Thompson, 1941). Mature trees which have low branches that are far from the ground may be relatively spared for leaf spot (Waterman, 1954b). Given that sexual phases of *Septoria* spp. may form on overwintering leaves on the ground, trees that grow less well in the plantation may be more susceptible to infection given that their leaves and branches are lower to the ground where the fungal inoculum exists.

We sought to determine obvious associations of precipitation and temperature with the frequencies of *Septoria* spp. detection among this and other studies. There is difficulty with the latter exercise since each study was approached differently. Multiyear studies with standardized approaches to testing and tabulation would be more appropriate.

Our findings of *S. musiva* in cankers support the work of Callan et al. (2007) and Beauseigle et al. (2010). Although *S. musiva* was isolated from some cankers, we must be careful to absolutely ascribe a cause and effect relationship. If *S. musiva* was plentiful in this and nearby plantations, it is possible that *S. musiva* surface contamination could lead to culture isolation even though this fungus is not the primary cause of disease. It is well known that cankers, even if caused by *S. musiva*, often have other fungi (e.g., *Cytospora*, *Dothichiza*, *Fusarium*, *Phomopsis*) identified (Waterman, 1946; Palmer et al., 1980; Ostry and McNabb, 1985). For argument against the latter, *S. musiva* was isolated from canker samples in this study after surface sterilization. Also, given the slow growth of *S. musiva* and its unlikelihood to overgrow other fungi, isolation implies some degree of purity. As is used in many other plant pathology studies, it would be of value however to directly identify *S. musiva* in diseased plants. This would require either direct staining techniques (e.g., direct immunofluorescence) or molecular studies (e.g., in situ hybridization). Such direct identification methods have not been cited in the scientific literature but are worthy of future study. It may not be surprising that there was no association of *Septoria* spp. on leaves with canker-like lesions in this study since there was no definitive evidence or systematic search from the affected trees to prove the causation of canker by fungus. It would be of value to identify *S. musiva*-cankered trees and then to determine the frequency of *Septoria* spp. on leaves from these same leaves.

S. musiva is capable of devastating tree plantations of poplars. If the fungus has become endemic in this province, even if only in a small region, the new introduction has the potential to be sentinel for further spread. Beauseigle et al. (2010) found this poplar pathogen on trees from natural stands in the vicinity of the plantation region assessed in this study. Search for further migration of the fungus from this focus is important. The potential threat to British Columbia plantations where susceptible seedlings or cuttings may be planted is evident since young plants are more likely to succumb to infection in contrast to old trees (Long et al., 1986). LeBoldus et al. (2007) found that disease variability was more likely linked to poplar genotype rather than fungal isolates.

Univariate analyses as conducted in this field study are exploratory at best. It would be of interest to furthering the science in this field by conducting multivariate analyses of *Septoria* spp. infection among black cottonwood. For example, susceptibility to infection might consider variables such as provenance, age of tree, location to other infected trees, consideration of flooding, tree height, diameter at breast height, intertree planting distances, among others. Furthermore, it is most likely that such studies will require multiyear investigation given the variable dynamics of pathogen, tree, and climate/environment.

Acknowledgements

I thank Stephanie Beauseigle for performing the molecular analytic work. I acknowledge the assistance of Drs. H. Kope and S. Zeglen (Victoria), Dan Carson (Kruger), and Dr. Y. El-Kassaby, for providing background to this study. I thank Dr. R. Hamelin for his tutelage in the planning and completion of this study.

References

Beauseigle S, Feau N, Hamelin RC. (2010) Poplar leaf spot and canker caused by *Septoria musiva* in British Columbia: risk assessment. Report to Pest and Forest Management, Province of British Columbia, April, 2010.

Bier JE. (1939) *Septoria* canker of introduced and native hybrid poplars. Can J Res [C] 17:195-204.

Bier JE. (1942) Forest pathology in British Columbia. Pulp Paper Magazine Canada 43:528-530.

Bier JE. (1961) The relation of bark canker to the development of canker diseases caused by native, facultative parasites. VI. Pathogenicity studies of *Hypoxylon pruinatum* (Klotzsch) Cke. and *Septoria musiva* Pk. on species of *Acer*, *Populus*, and *Salix*. Can J Bot 39:1555-1561.

Boes TK, Strauss SH. (1994) Floral phenology and morphology of black cottonwood, *Populus trichocarpa* (Salicaceae). Amer J Bot 81:562-567.

Callan BE. (1998) Diseases of *Populus* in British Columbia: a diagnostic manual. Cdn For Serv, Natural Resources Canada.

Callan BE, Leal I, Foord B, Dennis JJ, van Oosten C. (2007) *Septoria musiva* isolated from cankered stems in hybrid poplar stool beds, Fraser Valley, British Columbia. Pac NW Fungi 2:1-9.

Cellerino GP. (1999) Review of fungal diseases in poplar. FAO Corporate Document repository, Food and Agriculture Organization of the United Nations, Rome. Accessed in: http://www.fao.org/docrep/004/ac492e/ac492e00.htm (November, 2010)

Cimolai T. (2011) *Septoria* spp. disease and black cottonwood (*Populus trichocarpa*) in western North America: a review with special emphasis on *Septoria musiva*. Submitted as an Appendix to this thesis.

Cooper DT, Filer Jr. TH. (1976) Resistance to *Septoria* leaf spot in eastern cottonwood. Plant Dis. Rep. 60:812-814.

Dunlap JM, Braatne JH, Hinckley TM, Stettler RF. (1993) Intraspecific variation in photosynthetic traits of *Populus trichocarpa*. Can J Bot 71:1304-1311.

Dunlap JM, Heilman PE, Stettler RF. (1995) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. VIII. Leaf and crown morphology of native *P. trichocarpa* clones from four river valleys in Washington. Can J For Res 25:1710-1724.

Feau N, Weiland JE, Stanosz GR, Bernier L. (2005) Specific and sensitive PCR-based detection of *Septoria musiva*, *S. populicola* and *S. populi*, the causes of leaf spot and stem canker on poplars. Mycol Res 109:1015-1028.

Forest Pathology Herbarium, Pacific Forestry Centre, Victoria, B.C. http://www.pfc.cfs.nrcan.gc.ca/biodiversity/herbarium/index_e.html

Gornall JL, Guy RD. (2007) Geographic variation in ecophysiological traits of black cottonwood (*Populus trichocarpa*). Can J Bot 85:1202-1213.

Heilman PE, Stettler RF. (1985) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. II. Biomass production in a 4-year plantation. Can J For Res 15:384-388.

Howe GT, Hackett WP, Furnier GR, Klevorn RE. (1995) Photoperiodic responses of a northern and southern ecotype of black cottonwood. Physiol Plant 93:695-708.

Jones RK, Benson DM. (2001) Diseases of Woody Ornamentals and Trees in Nurseries. APS Press, The American Phytopathological Society, St. Paul, Minnesota.

LeBoldus JM, Blenis PV, Thomas BR. (2007) Evaluating the interaction between genotype and water stress in the hybrid poplar – *Septoria musiva* pathosystem. Can J Bot 85:1098-1102.

Long R, Bowersox TW, Merrill W. (1986) Artificial inoculation of *Populus* hybrids with *Septoria musiva*. Can J For Res 16:405-407.

Newcombe G, Chastagner GA, Callan BE, Ostry ME. (1995) An epidemic of *Septoria* leaf spot on *Populus trichocarpa* in the Pacific Northwest in 1993. Plant Dis 79:212.

Ostry ME, McNabb HS. (1985) Susceptibility of *Populus* species and hybrids to disease in the North Central United States. Plant Dis 69:755-757.

Ostry ME, Wilson LF, McNabb HS, Moore LM. (1989) A Guide to Insect, Disease, and Animal Pests of Poplars. United States Dept Agric, For Serv, Agriculture Handbook 677.

Palmer MA, Schipper Jr AL, Ostry ME. (1980) How to identify and control *Septoria* leaf spot and canker of poplar. Technical reference, North Central For Exp Stn, St. Paul, MN.

Riemenschneider DE, McMahon BG, Ostry ME. (1994) Population-dependent selection strategies needed for 2-year-old black cottonwood clones. Can J For Res 24:1704-1710.

Royle DJ, Ostry ME. (1995) Disease and pest control in the bioenergy crops poplar and willow. Biomass Bioenergy 9:69-79.

Sinclair WA, Lyon HH. (2005) Diseases of Trees and Shrubs, Second Edition. Cornell University Press, Ithaca, NY.

Smith JHG. (1957) Some factors indicative of site quality for black cottonwood (*Populus trichocarpa* Torr. & Gray) J Forest 55:578-580.

Strobl S, Fraser K. (1989) Incidence of *Septoria* canker of hybrid poplars in eastern Ontario. Can Plant Dis Surv 69:109-111.

Thompson GE. (1941) Leaf-spot diseases of poplars caused by *Septoria musiva* and *S. populicola*. Phytopathology 31:241-254.

Waterman AM. (1946) Canker of hybrid poplar clones in the United States, caused by *Septoria musiva*. Phytopathology 35:148-156.

Waterman AM, Aldrich KF. (1952) Surface sterilization of poplar cuttings. Plant Dis Rep 36:203-207.

Waterman AM. (1954a) Surface sterilization of hybrid poplar cuttings. Forest Research Notes, 32.

Waterman AM. (1954b) *Septoria* canker of poplars in the United States. United States Department of Agriculture, Circular No. 947.

Weber JC, Stettler RF. (1981) Isoenzyme variation among ten populations of *Populus trichocarpa* Torr. et Gray in the Pacific Northwest. Silv Genet 30:82-87.

Weber JC, Stettler RF, Heilman PE. (1985) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. I. Morphology and phenology of 50 native clones. Can J For Res 15:376-383.

White TJ, Bruns T, Lee S, Taylor J. (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. *In*: Innis MA, Gelfand DH, Sininsky JJ, White TJ eds. PCR Protocols: A Guide to Methods and Applications. Academic Press, San Diego, USA. pp. 315-322.

Zalasky H. (1978) Stem and leaf spot infections caused by *Septoria musiva* and *S. populicola* on poplar seedlings. Phytoprotection 59:43-50.

Table 1. Comparison of historic average monthly high and low temperatures and historic average monthly precipitation of the Harrison Mills plantation to other regions of British Columbia. Temperatures are in Centigrade, and precipitation is expressed in millimetres. Data were obtained from Environment Canada and supplemented with data from The Weather Network and The Weather Channel (www.weatheroffice.gc.ca, www.theweathernetwork.com, www.weather.com, all accessed in February, 2011). Single letters represent chronological months of the year.

Average Precipitation (mm.)	J	F	M	A	M	J	J	A	S	O	N	D
Harrison Mills Plantation	231	175	150	119	97	81	61	58	99	175	241	241
Victoria	142	99	71	43	33	28	18	23	36	74	140	241
Prince Rupert	252	216	188	180	142	119	118	163	244	380	290	280
Prince George	53	36	36	28	51	64	61	61	58	58	53	53
Okanagan Valley (Kelowna)	31	23	23	25	38	38	33	33	36	23	28	38
Fort St. John	31	23	25	20	41	66	74	58	43	28	28	31

Average High Temperature (^{o}C)	J	F	M	A	M	J	J	A	S	O	N	D
Harrison Mills Plantation	4	8	11	14	18	21	23	23	21	14	8	5
Victoria	6	8	10	13	16	19	22	22	19	14	9	7
Prince Rupert	4	6	7	9	12	14	16	16	15	11	7	4
Prince George	-6	-1	4	11	16	19	22	21	16	9	1	-5
Okanagan Valley (Kelowna)	-1	3	9	15	20	24	27	27	21	13	5	5
Fort St. John	-11	-7	-1	8	15	19	21	20	14	8	-3	-9

Average Low Temperature (^{o}C)	J	F	M	A	M	J	J	A	S	O	N	D
Harrison Mills Plantation	-1	1	3	4	8	11	12	12	10	6	3	-1
Victoria	-1	1	2	3	6	9	11	11	8	5	2	1
Prince Rupert	-3	-1	0	1	4	7	9	10	7	4	1	-2
Prince George	-14	-11	-6	-2	3	6	8	7	3	-1	-7	-13
Okanagan Valley (Kelowna)	-9	-7	-3	4	4	8	10	9	5	2	-3	-7
Fort St. John	-19	-16	-11	-2	3	8	10	9	4	-1	-11	-18

Table 2. Frequencies of *S. musiva* and *S. populicola* detection compared among this study in 2010 and that of Beauseigle et al. (2010) from 2008 and 2009. Data for 1993 have been added to the Table given that it was declared by Newcombe et al. (1995) that epidemic leaf spot attributed to *S. populicola* was found among natural black cottonwood; these data included samples from British Columbia. Average temperature (Centigrade) per month and average rainfall (millimeters) per month are noted for timings prior to sampling of leaves for leafspots in each study; leaves were sampled in late September to early October for each year. Single letters represent chronological months of the year from April to August. P = black cottonwood provenance stand, FVpop = Fraser Valley natural poplars outside of plantation area, HM = other non-provenance trial Harrison Mills plantation.

Year	% *S. musiva*			% *S. populicola*			months (rainfall/temperature)					average rainfall			average temperature		
	P	FVpop	HM	P	FVpop	HM	A	M	J	J	A	early 3 mo. leaf growth	prior 2 mo.	prior 5 mo.	early 3 mo. leaf growth	prior 2 mo.	prior 5 mo.
2010	6.4	-	-	70.1	-	-	110/9.7	107/12.4	83/15.5	3/18.9	43/18.8	100	23	69	12.5	18.9	15.1
2009	23.4	1.3	-	32.5	62.1	-	94/10.0	139/13.5	23/18.0	23/20.8	42/19.0	85	33	64	13.8	19.9	16.3
2008	0	1.2	33	25	64.4	24	79/7.3	106/14.7	74/14.9	57/18.5	215/19.1	86	136	106	12.3	18.8	14.9
1993	-	-	-	‘epidemic’ *S. populicola*			145/10.3	103/16.1	118/16.0	59/17.2	47/18.3	122	53	94	14.1	17.8	15.6

Table 3. Frequencies of *S. musiva* and *S. populicola* among black cottonwood provenances (% in brackets).

Provenances	South Coast	USA	Mid-Coast	Prince George	Smithers	Dease Lake	Far North
Total trees sampled	155	24	48	24	34	12	3
***S. musiva* positive**	6 (3.9)	2 (8.3)	5 (10.4)	1 (4.2)	3 (8.8)	2 (16.7)	1 (33.3)
***S. populicola* positive**	102 (65.8)	16 (66.7)	39 (81.3)	16 (66.7)	29 (85.3)	8 (66.7)	2 (66.7)

Table 4. Frequency of *S. musiva* isolation from provenance clusters (% in brackets).

	***S. musiva* isolation**	
provenance	positive	negative
South Coast/USA	8 (4.5)	171
Other	12 (9.9)	109

Table 5. Frequency of *S. populicola* isolation from provenance clusters (% in brackets).

	***S. populicola* isolation**	
provenance	positive	negative
South Coast/USA	118 (65.9)	61
Other	94 (77.7)	27

Table 6. Frequency of *S. musiva* isolation from trees with or without canker-like lesions (% in brackets).

	***S. musiva* isolation**	
	positive	negative
with lesion	5 (8.5)	54
without lesion	14 (5.9)	224

Table 7. Frequency of *S. populicola* isolation from trees with or without canker-like lesions (% in brackets).

	***S. populicola* isolation**	
	positive	negative
with lesion	40 (67.8)	19
without lesion	170 (71.4)	68

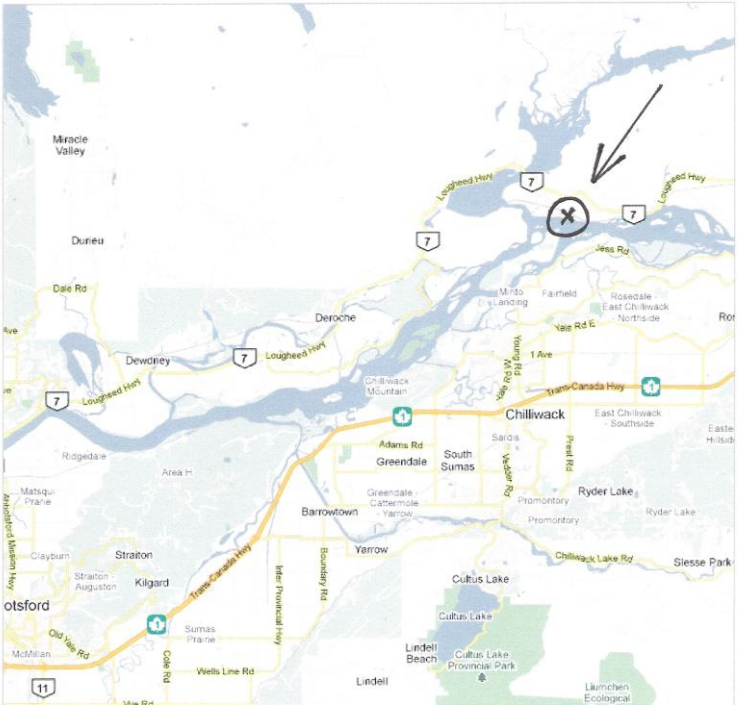
Table 8. Frequency of *S. musiva* isolation from trees with either stem or branch canker-like lesions (% in brackets).

	***S. musiva* isolation**	
	positive	negative
stem lesion	1 (7.1)	13
branch lesion	4 (8.5)	43

Table 9. Frequency of *S. populicola* isolation from trees with either stem or branch canker-like lesions (% in brackets).

	***S. populicola* isolation**	
	positive	negative
stem lesion	8 (57.1)	6
branch lesion	33 (70.2)	14

Figures 1 and 2. Geographic location of the black cottonwood plantation assessed in this study. Note immediate proximity to Chilliwack, British Columbia (courtesy of Google Maps, Canada).



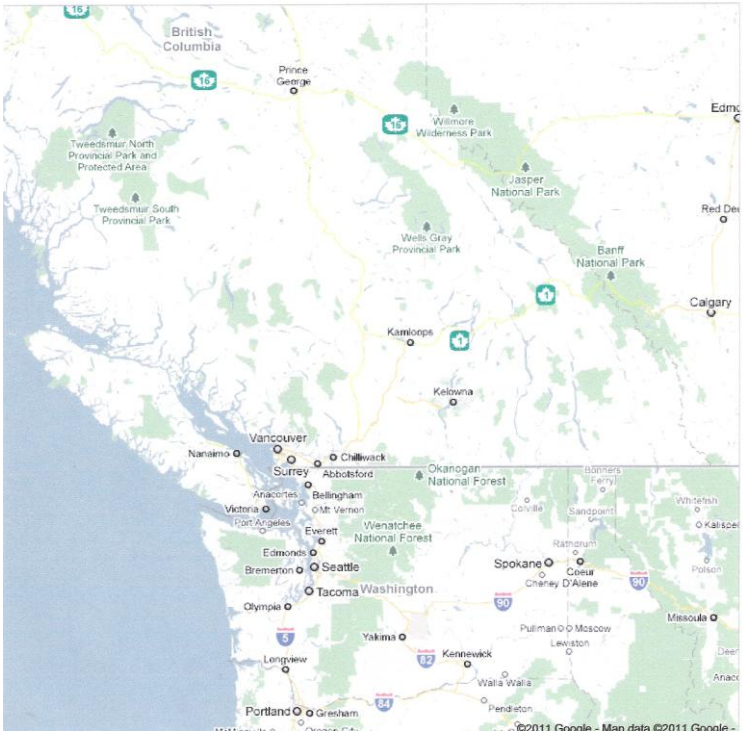


Figure 3. Sources of *P. trichocarpa* provenances from the Harrison Mills plantation. Note the acquisition from a few sources in Washington State and Oregon (obtained from Dr. Harry H. Kope, Forest Practices and Investment Branch, British Columbia, Ministry of Forests, Mines, and Lands, Victoria, British Columbia).

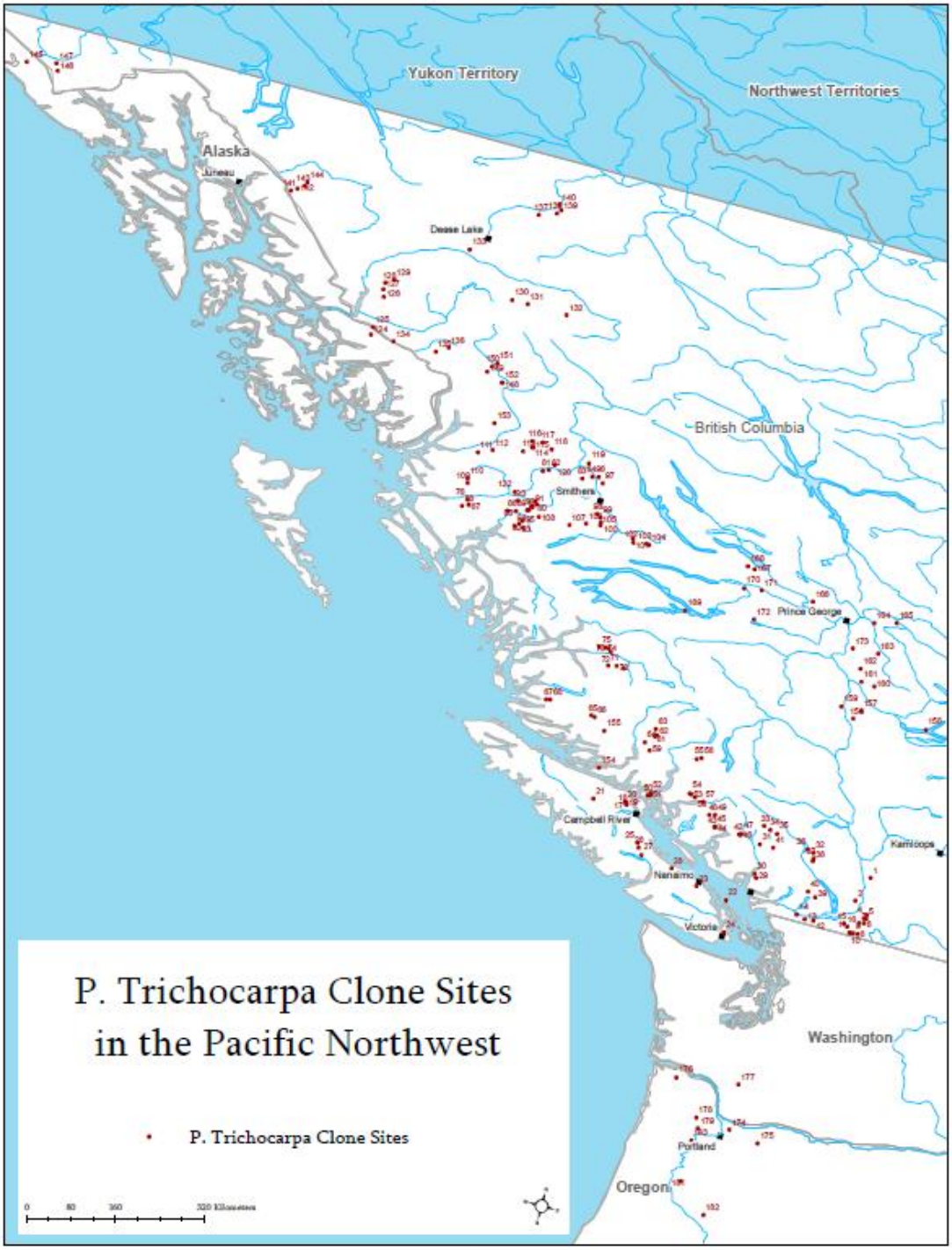


Figure 4. Early stages of a stem bark canker with some invasion. Note leaf spots in background leaves (photo – Tomas Cimolai, September, 2010).

Figure 5. A large stem canker which phenotypically has affected a small branch, but which has not yet considerably affected the distal growth. Such a lesion would be very susceptible to cracking after pressure from wind and natural elements. Note leaf spots or similar among background leaves (photo – Tomas Cimolai, September, 2010).

Figure 6. A stem canker having encircled the entire primary stem. A secondary stem may be emerging which could allow the tree to survive. The main stem canker is causing tree fall. Note classic *Septoria* leaf spots in some background leaves (photo – Tomas Cimolai, September, 2010).

Figure 7. A large stem canker which has invaded the tree considerably. The tree is imminently susceptible to tree fall. In such a canker, regardless of the primary cause, it is likely that secondary saprophytic fungi are co-infecting. The combination of primary and secondary stem pathogens eventually contributes to this appearance (photo – Tomas Cimolai, September, 2010).

Figure 8. A stem canker which involves approximately 50% of the circumference. The outer edges of the canker as well as the top and bottom ends appear to be lignifying. Such change in the canker may eventually be associated with tree healing and cessation of further disease and decay (see Cimolai, 2011). Note leaf spots in background leaves (photo – Tomas Cimolai, September, 2010).

Septoria spp. Disease and Black Cottonwood (*Populus trichocarpa*) in Western North America: A Review With Special Emphasis on *Septoria musiva*

(submitted as an Appendix to B.S.F. thesis entitled "*Septoria* spp. Infection Among North American Northwest Coast Provenances of Black Cottonwood (*Populus trichocarpa*): A Field Study of Natural Infection in a Fraser Valley Plantation")

Contents

I. Introduction

Poplars are widespread in forests across the Northern Hemisphere, and they have been integral to mankind for thousands of years (Gordon, 2001). Canada is home to 7 of the 29 naturally occurring *Populus* species and many introduced artificial hybrid strains (Cooke and Rood, 2007). *Populus* species and their natural hybrid strains are vitally important in their local ecological roles. Canada has had a long and strong tradition in studying poplars (Richardson et al., 2007).

Poplars are synonomous with rapid development, ease of dissemination, and wide ecological amplitudes. Such characteristics have contributed to their popularity historically, and they now are of increasing importance given the prevalence of climate change. In British Columbia, *Populus trichocarpa* (black cottonwood) is native and is largely utilized in industrial fibre production plantations especially from hybrid trees. The significance of *Populus* species in British Columbia continues to increase, given their potential to act in evolving roles such as bioenergy, carbon sequestration, phytoremediation, riparian rehabilitation, and wood supply. The latter is consistent with the growing importance of poplar plantations for commercial purposes worldwide (Sedjo, 2001).

Populus trichocarpa in British Columbia is susceptible to *Septoria* spp. fungi either producing leaf spot or inducing cankers (Newcombe et al., 1995). *Septoria* spp. have the potential to impede growth and cause mortality among black cottonwood. Although *S. musiva* in particular is commonly found causing disease in eastern North America, such a relationship with poplars also exists in the Fraser Valley of British Columbia (Callan et al., 2007). The significance of *Septoria musiva* in British Columbia

is not fully understood. This paper investigates the relationship between *Populus trichocarpa* and *Septoria* spp. in British Columbia. The potential role for *S. musiva* is highlighted in a scenario where it is feared that a large scale spread could occur throughout the province. Such an occurrence could result in widespread disease for *Populus trichocarpa*, which would jeopardize dependent ecosystems and dependent industrial forestry operations. This review initially examines the biology of *P. trichocarpa* and then follows with a comprehensive review of *Septoria* spp. disease as related to this and similar poplars.

II. Overview of Black Cottonwood (*Populus trichocarpa*)

i. History of *Populus trichocarpa*

The *Populus* genus can be traced through fossil records which possibly date the appearance of these trees back to the late Paleocene or early Eocene periods (~50-60 million years ago) (Slavov and Zhelev, 2010). *Salix* (willow) hardwoods have been found in fossil records to precede *Populus*, but both are said to be among the earliest of flowering plants (Boes and Strauss, 1994). There is controversy as to how *Salix* and *Populus* may have diverged or may have been derived from one another. Overall, *Populus* seems to be the more primitive of the two and *Salix* has become more diverse (Dickmann and Kuzovkina, 2008). Precursors of the current *Populus* Sections are believed to have been available by the Miocene period. During these times, primitive non-*Tacamahaca* Sections were further evolved, and subsequent *Tacamahaca* development arose later (some say late Oligocene). Natural hybridization, genetic diversity, and adaptation and selection then led to the range of *Populus* now encountered

worldwide. Indeed, the current understanding of the taxonomy is complicated because some believed that natural hybrids constituted new species, although some resolution in this regard has now been proposed so as to have fewer species in fewer Sections (Slavov and Zhelev, 2010). This history is relevant because more closely related *Populus* spp. hybridize readily, whereas some potential crosses cannot be accomplished. Despite what should have been considerable opportunity to diversify over millions of years, structural diversity in this genus is limited (Boes and Strauss, 1994).

In a general sense, *Populus* spp. have been associated with human civilizations and their riparian ecologies throughout the world (Stanton, 2009). In this context, the trees have been harvested for lumber, heat, biomass, forage, wind protection, and shade. Details of poplar use can be found in records of ancient Roman and ancient China. New technology has opened the door for poplar use in the modern era

P. trichocarpa is a deciduous broadleaf tree that is native to the northwestern region of North America. It is variably known by its synonyms as black cottonwood, western balsam poplar, balsam cottonwood, and California poplar. The following scientific names are considered synonomous with *P. trichocarpa*:

P. balsamifera subsp. *trichocarpa*

P. balsamifera var. *californica*

P. hastata

P. trichocarpa subsp. *hastata*

P. trichocarpa var. *hastata*

P. trichocarpa var. *cupulata*

P. trichocarpa var. *ingrata*

The scientific classification of this species is as follows:

Kingdom	Plantae
Division	Magnoliophyta
Class	Magnoliopsida
Order	Magpighiales
Family	Salicaceae
Genus	*Populus*
Section	*Tacamahaca*
Species	*Populus trichocarpa*

The type strain is known as *P. trichocarpa* Torrey & Gray. It is regarded as the largest of hardwood trees in the Pacific Northwest (DeBell, 1990). The Salicaceae Family includes willows and poplars, and they are common throughout the world, but *P. trichocarpa* is indigenous as a 'pioneer' species to the Pacific Northwest of North America. The Genus *Populus* includes poplars, the name of which is derived from the Latin "[trees] of the people" (Gordon, 2001). The Genus *Populus* has been divided into six sections, the trees of which are spread throughout the world (Table 1). Slavov and Zhelev, 2010) It is estimated that *Populus* species comprise ~10% of the Salicaceae Family (Boes and Strauss, 1994). *P. trichocarpa* is assigned to the section *Tacamahaca*. Some believe that the species *P. balsamifera* and *P. trichocarpa* are considerably similar, and it has been suggested that *P. trichocarpa* could be classified as *P. balsamifera* subsp. trichocarpa (Dickmann and Kuzovkina, 2008). The origin of 'trichocarpa' is Greek and is a composite of 'tricho' which means hair and 'carpa' which means fruit. The latter

Table 1. Taxonomic sections of the Genus *Populus* (as per Slavov and Zhelev, 2010).

Section	Species	Geographic locale
Abaso	*P. mexicana*	Mexico
Turanga	*P. euhratica*	Africa, Asia
	P. ilicifolia	Africa
	P. pruinosa	Asia
Leucoides	*P. glauca*	China
	P. heterophylla	United States
	P. lasiocarpa	China
Aigeiros	*P. deltoides*	North America
	P. fremontii	United States
	P. nigra	Eurasia, Africa
Tacamahaca	*P. angustifolia*	North America
	P. balsamifera	North America
	P. ciliata	Himalayan region
	P. laurifolia	Eurasia
	P. simonii	Asia
	P. suaveolens	Japan, China
	P. szechuanica	Eurasia
	P. trichocarpa	North America
	P. yunnanensis	Eurasia
Populus	*P. adenopoda* Maximowicz	China
	P. alba	Europe, Africa, Asia
	P. gamblei	Eurasia
	P. grandidentata	North America
	P. guzmanantlensis	Mexico
	P. monticola	Mexico
	P. sieboldii	Japan
	P. simaroa	Mexico
	P. tremula	Europe, Africa, Asia
	P. tremuloides	North America

described the flowering stage of the tree. Natural hybrids of *Populus* and *P. trichocarpa* can occur in nature, but most hybrids that incorporate *P. trichocarpa* genetics are experimentally derived.

The genome of *P. trichocarpa* has now been sequenced (Tuskan et al., 2006; Douglas and DiFazio, 2010). The genome size is approximately 485 million base pairs,

consists of 19 chromosomes, and maintains some 45,000 or more genes. In addition, there is also a separate mitochondrial genome of 803,000 base pairs (52 genes) and a separate chloroplast genome of 157,000 base pairs (101 genes). In contrast, the human genome includes nearly three billion base pairs.

ii. Distribution and typical climate

P. trichocarpa is predominantly found in the west/northwest of North America and especially in coastal aspects of the continent. On one hand, the distribution is as far north as 62^{o} North from Cook Inlet; it then extends into southeast Alaska and into British Columbia (Figures 1 and 2). In the main geography of the United States, the tree is found in forested, especially coastal, Washington and Oregon and then extends to the mountains

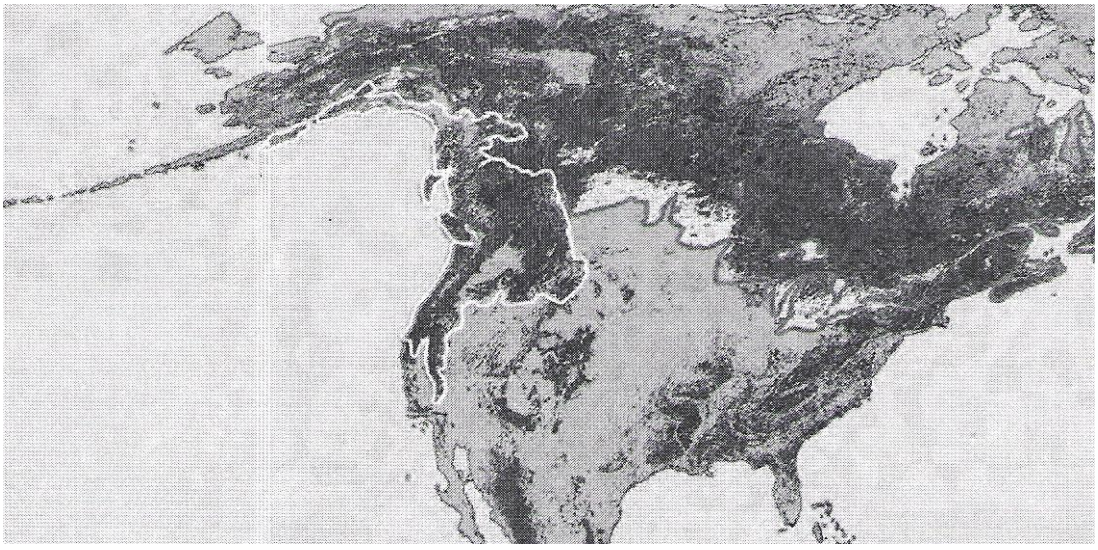


Figure 1. Distrubution of *P. trichocarpa* in North America. The major area of distribution is outlined by the white border in the Pacific Northwest (from Dickmann and Kuzovkina, 2008).

Figure 2. Distribution of *P. trichocarpa* in British Columbia. Note the 'very frequent' density in the multitude of riparian systems in this province (from Klinka et al., 2000).

of California and northern Baja California. This poplar can also be found in some other regions in B.C. west of the Rockies and occasionally in western aspects of Alberta and

Montana and north Idaho. Small clusters are also found in the southeast of Alberta, Montana east, west in North Dakota, west in Wyoming, Utah, and Nevada.

The locale of the black cottonwood is often referred to as 'riparian' meaning along the banks, flood plains, and islands of rivers and streams (de Vries, 2001; Dickmann and Kuzovkina, 2008). These areas are alluvial, morainal, and outwash habitats which are moist and generally wetlands with a higher water table and periodic seasonal flooding. There is often sand and silt where water is recessing from gravel. Such sites often change, and the ecosystem may link running and standing water, may link aquatic domain with terrestrial areas, and may interface ground water with surface water. The best growth occurs proximal to the moist Pacific Ocean near the coast. As well by the coast, the trees may be found up to 1500 metres above sea level, whereas inland they may be found up to altitudes of 2000 metres above sea level in canyons and valleys. Such distribution is critical to the trees since they have relatively high water consumption. Apart from the necessary association of *P. trichocarpa* with such humid terrains, one may also see this tree and other similar as being a source for preserving the environment and perhaps even restoring the ecosystem when otherwise stressed (de Vries, 2001) .Klinka et al. (2000) described the presence of black cottonwood in the following zones for British Columbia: more so in SBS, ICH, and CWH, lesser in IDF and CDF, and least in lower MH, lower ESSF, MS, BWBS, SBPS, BG, and PP.

In particularly analyzing the distribution of *P. trichocarpa* in British Columbia, Smith (1957) in his classical studies concluded that the distribution of the tree would be favoured in bare and moist mineral soils which were exposed to seedfall in the early summertime; the exposure of such soil could best occur close to rivers and streams in the

fresh alluvial deposits. He also observed that *P. trichocarpa* would occur early in these regions but would eventually be replaced by more tolerant grasses, shrubs, and trees. In logged areas, black cottonwood would be replaced by less desirable trees and brush. As for soil requirements, Smith found that nutrients, soil oxygenation, and pH 6-7 were beneficial in addition to moisture. A layer of loam or heavy soil greater than one foot in depth was essential if the base had gravel underlay. Although stagnant water was a detriment, flooding with fast-moving water could still allow adequate oxygenation and the net inflow of sediment nutrients. Clay soils were supportive. The adaptation of this tree is illustrated in the Figure 3 as arranged by Klinka et al. (2000).

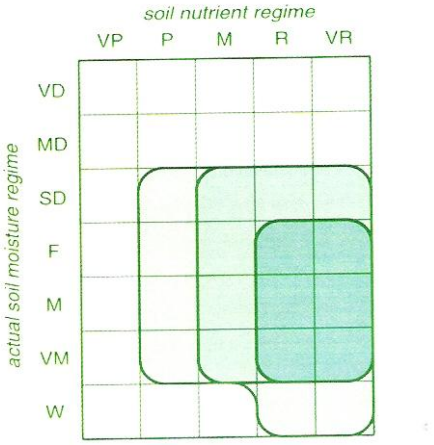


Figure 3. Diagram illustrates the general requirements of *P. trichocarpa* in regards to soil nutrients and soil moisture. Soil nutrients are rated as VP – very poor, P – poor, M – medium, R – rich, and VR – very rich. Soil moisture is rated as VD – very dry, MD – moderately dry, SD – slightly dry, F – fresh, M – moist, VM – very moist, and W – wet (from Klinka et al., 2000).

Black cottonwood is drought intolerant and generally shade intolerant. The trees can be susceptible to extremes of heat (not usually found where summer temperatures excess the high 30^{o}C for over one week) and cold (not usually found where temperatures can exceed lower than -40^{o}C; excess cold can cause trees to crack due to high moisture content) (DeBell, 1990). Although *P. trichocarpa* is found growing well in the Pacific Northwest where rainfall is heavy in total for the year, most of the tree growth, however, occurs during the period when only 1/3 of the total rainfall occurs.

iii. Seed production and fruiting bodies

P. trichocarpa is said to be dioecious, that is, female and male reproductive structures exist on separate trees (DeBell, 1990). Although the female/male ratio of trees is generally even, different environments may favour a skew of this ratio in either direction. The flowering reproductive structures are commonly referred to as 'catkins', and these are pendulous and pendunculated as they hang from the tree (Figures 4 and 5). Flowers emerge before the leaves emerge; poplars are among the earliest of flowering plants in the spring (Slavov and Zhelev, 2010). In British Columbia, the catkins are formed as early as the beginning of March and continue to be seen until approximately mid-June; these timings will be later in *P. trichocarpa* specimens from interior B.C. and northern B.C. Trees will not commonly produce these flowering structures until they reach nearly 10 years in the wild, although trees in plantation may flower earlier (Slavov and Zhelev, 2010). Compared to other poplars, *P. trichocarpa* has a larger number of reproductive structures per tree (Boes and Strauss, 1994).

Figure 4. Photograph illustrates a male catkin and some tree buds (from http://en.wikipedia.org/wiki/File:Black_Cottonwood_male_catkin_and_leaf_buds.jpg).

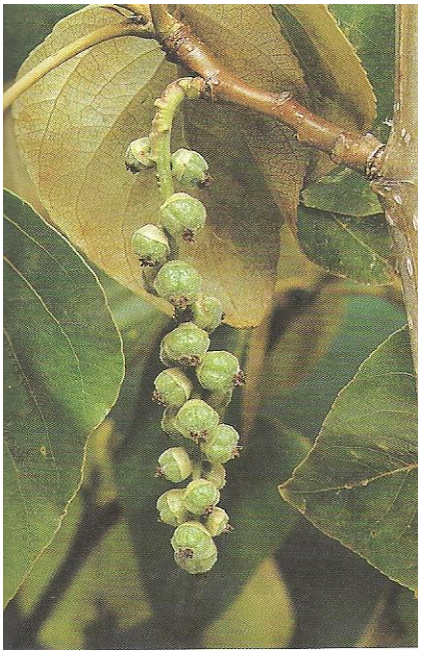


Figure 5. Photograph illustrates three carpel fruiting bodies (from Farrar, 1995).

Male/female flowering differentiation will take place by late spring. Male catkins bear some 30-60 stamens per flowering structure. The male catkins are 2-3 cm. long, and the flowering structure is deciduous. The female structures bear 3 stigma per flower and may be as long as 8-20 cm. When the stigma receives pollen, fertilization occurs within a day or so. The subsequent fruit is round, is about 5-8 mm. long, and has three carpels, occasionally four. In the early stages of flower formation, some find *P. trichocarpa* to be indistinguishable from *P. balsamifera*. *P. trichocarpa* has more stamens, and the fruit has 3 (or 4) carpels compared to the 2 of *P. balsamifera* (Dickmann and Kuzovkina, 2008). The capsule will contain some 30-50 seeds each, and the seeds resemble long white cottony hairs (Boes and Strauss, 1994;Pojar and MacKinnon, 1994). Histological detail of organogenesis is beautifully illustrated by Boes and Strauss (1994).

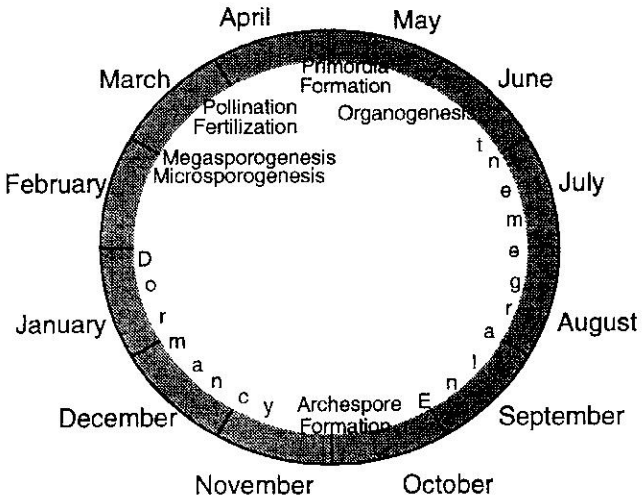
The seed capsules will dehisce in 4-6 weeks after fertilization, and literally millions of seeds can be sourced to one tree. The seeds remain viable for 1-2 weeks, and when reaching a viable site, germination will require only 24 hours.

A typical cycle of *P. trichocarpa* phenology is illustrated by Figure 6 which is derived from trees in Oregon, but which would be generally similar to that in British Columbia (Boes and Strauss, 1994).

iv. Seed dissemination and early growth

The seeds are disseminated during late May to July via wind and water. The seeds are considerably light so as to facilitate distant transport. Although typical seed viability may be for two weeks or so, less often germination can occur up to one year later under the appropriate conditions (DeBell, 1990). Seeds tend to be relatively hardy,

Figure 6. Typical phenology as observed for *P. trichocarpa* in Oregon (Boes and Strauss, 1994).



and there is a high rate of germination, but the majority of sprouts will die; some estimate ~75-100% die off under conditions of drying, excessive flooding, and many other soil disturbances, let alone the susceptibility of young growth to animals (Slavov and Zhelev, 2010). The ability of *P. trichocarpa* to arise from seeds is evidently a follow-through to the sexual reproductive capabilities, but this species, in common with other Section *Tacamahaca* cottonwoods, is very capable of propagation through asexual means (DeBell, 1990).

Asexual reproduction occurs through various modes. Root sprouts may turn into fully viable trees. Shoots may arise from broken branches with subsequent rooting. Entire tree trunks or large branches that are toppled and severed may become covered with sediment and thereafter establish shoots and roots. Different parts of the tree may be sheared with the forces that occur with stream and river flooding and movement in the

riparian system. An excellent example of these phenomena was described by Rood et al. (2003a) who studied black cottonwood cyclical patterns in the Elk River district of British Columbia (near the Crowsnest Pass, southeastern province). Examining the Elk River floodplain after a record flood in 1995, they determined that 91% of saplings were established from sheared branches, 7% from beaver cuttings, and only 1% appeared to have arisen from branch plantings (cladoptosis). Even under greenhouse conditions, only 3% of abscised branches were successfully propagated in contrast to 96% success with winter shoot cuttings. In the first year of natural early growth, saplings from branch fragments were much more hardy than seedlings directly derived from seeds (Figures 7 and 8). Figure 9 illustrates the amount of debris that can arise with tree destruction, shearing, and other physically impactful activities.

Figure 7. Multi-stemmed *P. trichocarpa* sapling growing from a rooted branch and contrasted to growth from seed sprouting (from Rood et al., 2003a).

Figure 8. Rooted branch fragment of *P*. *trichocarpa* with several shoots contrasted to growth from seed sprouting (from Rood et al., 2003a).

Figure 9. A riparian zone along the Elk River, British Columbia showing healthy black cottonwood as well as woody debris arising from the flood conditions (from Rood et al., 2003a).

As an extension from the above, trees may be propagated by the purposeful acquisition of stem and shoot cuttings (Dickmann and Kuzovkina, 2008). Sprouts may also arise from cut stumps (coppicing). Figure 10 illustrates the growth that can arise from cuttings. Smith et al. (1956) examined the growth of cuttings from *P. trichocarpa*. They found that there was a tendency for the growth from a cutting to maintain characteristics of the position from where the cutting was acquired from the parent tree. The position of cutting from tree crown influenced growth height and cutting survival. They established that long cuttings from one-year leaderwood was preferable. Better growth was obtained from leaders than secondary branches. Figure 11 illustrates the phenomenon of root suckering.

Figure 10. Root propagation from a stem cutting of *P. trichocarpa* (from Stanton, 2009).

Figure 11. Root suckering phenomenon in cottonwoods (from Stanton, 2009).

v. General growth characteristics

Populus spp. in general have high photosynthetic and carbon uptake potential (Dillen et al., 2010). They have good leaf area development, sylleptic branches, phenological adaptation, phytohormone regulation, among other important characteristics. *P. trichocarpa*, in particular, is the largest poplar and has the highest number of sylleptic branches (Ceulemans et al., 1992). The crowns are narrow, cylindrical, and round-topped (Dickmann and Kuzovkina, 2008). Roots are widespread. The tree has high levels of rooting hormone so that early sprouting can occur. For example, after logging, natural regeneration can take place with rooting of fragments that are buried or with stumps. These phenomena also give rise to the ability of the tree to rapidly resprout after cutting in coppice stands. Growth in general is dependent on both genetic and environmental factors (Mohn, 1969). Studies of the black cottonwood have occurred mainly in Washington State and British Columbia.

Seedlings of *P. trichocarpa* take best in barren soil where there is little competition. Lesser adaptation and growth will occur in underprepared soils or in old logging areas (DeBell, 1990). Logged area regeneration, however, is improved if the region is clearcut. Within the initial five years of growth, trees will be thinned out naturally as some cannot compete with more vigorous trees and with the shade that may develop. In British Columbia, very good pulpwood-ready trees can be obtained in about 10-15 years, and decent saw logs will be achieved by about 20-25 years. There can be up to a four-fold difference in biomass availability as measured in wood dry weight between the perceived best clones versus the perceived worst clones of the black cottonwood. When investigating provenances from Oregon, Washington, and British Columbia, the ten best biomass producing clones originated from six different provenances, thus indicating the wide variability of *P. trichocarpa* in nature (Heilman and Stettler, 1985a).

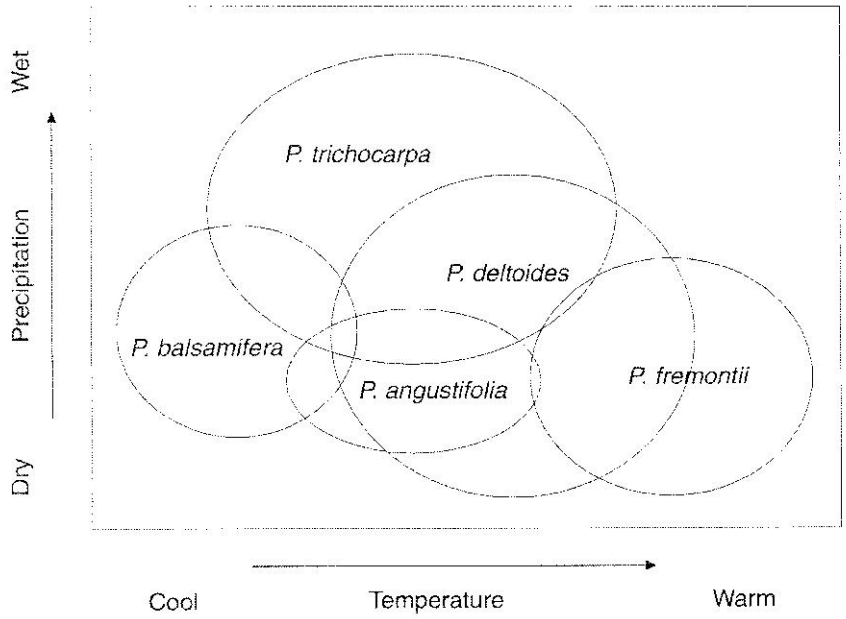
In the Lower Fraser Valley, planted *P. trichocarpa* averaged 8" diameter at breast height (dbh) and up to 55' tall after some ten years (DeBell, 1990). Some trees have been found to reach nearly 200 years of age in the wild, with large trees up to 72-120" dbh and 200' tall. In the Lower Fraser Valley, mean annual increments in wood are approximately 10-15 m^3/hectare. Generalizing for British Columbia, the best natural growth of black cottonwood will take place in the Lower Fraser Valley and the Squamish River Valley (Smith and Blom, 1966). Lesser growth is found in the Skeena, Upper Fraser, and Thompson River Valleys, and the worst growth is to be found in the dry interior valleys. Interior B.C. poplars are more likely to have defects and diseased growth, and thus there is a lower quality of this tree in that region. Fraser Valley islands are particularly suitable for supporting tree growth due to the intermittency of flooding which brings nutrients.

These trees are favoured by long frost-free periods, and they are susceptible to sudden frost. Inland clones exhibit greater frost tolerance and lesser winter kill than coastal trees. Defoliation appears to increase tree susceptibility to frost. There is considerable variation to frost tolerance among trees of the same provenance or region (McCamant and Black, 2000). *P. trichocarpa* is relatively wind tolerant.

Rood et al. (2003b) have discussed changes in riparian systems and their effect on cottonwood. In such systems, trees are dependent on shallow ground water. If the upstream is dammed or if the downstream water is depleted otherwise, the water table will drop to cause stress after a lag time. The introduction of irrigation waters can mitigate water deficiency. Drought responses in trees include stoma closure, decreased transpiration, decreased carbon uptake, decreased photosynthesis, decreased water potential, and the development of xylem cavitation. These changes in turn lead to decreased shoot growth, alterations in roots, loss of branches, die-back of the crown, and potentially tree mortality altogether. There is considerable variation in nutrient utilization efficiency, but *P. trichocarpa* is considered to have good efficiency in this regard overall; nutrient depletion over time, however, can be a limiting factor (Heilman and Stettler, 1986). Figure 12 illustrates general adaptive ranges of riparian cottonwoods in North America. If alluvial deposition is repetitive in the area, there may be up to 10 years variation in tree age on the same site. Estimates of growth in riparian regions of Chilliwack, B.C. previously found natural frequencies of up to 43 trees/acre with dbh greater than 11.5” and up to 136 trees/acre total black cottonwood (Smith, 1957).

Timing of natural spring growth is associated with changes in temperature in the river drainage areas while changes in the autumn are associated with both temperature

Figure 12. Adaptation of riparian cottonwoods in North America. Note the adaptation of *P. trichocarpa* to favour regions with greater precipitation (from Rood et al., 2003b).



gradient and disease pressures (Stanton, 2009). Northern provenances have better photosynthetic capability, although they are more photoperiod sensitive (Howe et al., 1995). Photosynthesis is better among trees from lower xeric environments compared to upper mesic environments of the same valley, but generalizing, mesic clones have better photosynthesis than xeric clones (Dunlap et al., 1993). Water use efficiency is better among trees from arid continental climates and lower in those from coastal areas. Seedlings can survive up to 6-8 weeks of flooding, but flooding will have an impact on decreasing branch length and leaf number (Smit, 1988). Resistance to seasonal flooding does not seem to be genetically determined.

British Columbian provenances have been examined in some detail (Gornall and Guy, 2007). Carbon dioxide assimilation increases with increasing latitude of the provenance but not among clones within that provenance. Stomatal density also correlated with latitude. Northern provenances have increased carbon dioxide assimilation and stomatal conductance to allow for the shorter growing season. The longevity of leaves is inversely related to latitude. Stem volume is associated with leaf area and leaf growth rate (Ridge et al., 1986). In Washington state, phenology variation has been detailed from different river valleys, both mesic and xeric (Dunlap et al., 1995; Dunlap and Stettler, 1996). Trees in mesic zones have larger size traits. Trees from eastern Washington (more xeric) have more water deprivation resistance than those from western Washington (Bassman and Zwier, 1991). As one moves from provenances in the southwest of Oregon and Washington to the northeast towards Washington-British Columbia, it is apparent that the southwest trees have smaller leaves and more numerous branches along with later fall growth (Weber et al., 1985). When comparing black cottonwood from southeastern British Columbia to that from Idaho, it was determined that leaf morphology was not a useful predictor for tree height (Riemenschneider et al., 1994). From studies of the latter, it was proposed that selection of favourable tree characteristics needed to be considered on an individual population basis. Nevertheless, there is plenty of variation even within the same region. For example, leaf size and other measures can be adapted even at a single riparian level along a single slope of forest (Dunlap et al., 1995). Looking at enzyme variation among black cottonwood from the Pacific Northwest, Weber and Stettler (1981) found an overall high genetic similarity among trees from different riparian populations, although there was still considerable

differences among clones from the same region. The regions with the most differences were those of large river drainages. Thus, it is apparent that *P. trichocarpa* has responded to climatic selection pressures at both local and regional levels.

Black cottonwood growth has also been well studied in plantations and greenhouses (DeBell, 1990). Cuttings are usually planted 12-16" deep. The cuttings are generally 16-24" long and 0.4-1.2" in diameter. There is better growth when cuttings have pre-existing buds. Short cuttings are less susceptible to crooked growth later (Heilman and Ekuan, 1979). Cuttings can yield 5 feet of growth in one year and up to 20 feet in four years. Spacing of trees at 6' by 6' gave increased yield of wood compared to larger spacing of trees (Smith and Blom, 1966). Greater spacing was associated with decreased yield perhaps due to greater competition secondary to weed pressure. "Sets" are recommended for planting in commercial scale plots. There appears to be considerable risk in cultivating pure *P. trichocarpa* because of the high risk for disease in plantations. Weed growth is a major detriment to growth in the first few years (Silen, 1947; DeBell, 1975). Fertilizers are often applied, and pulp mill sludge can be used for the latter, although it may slow growth for the first two years (DeBell, 1975). Growth in the first two years of the tree determines tree fate largely. Leaf and canopy features correlate with stem growth thereafter (Dunlap and Stettler, 1998). Clone to clone variability is high, however, and this gives rise to the potential for selecting better cultivars. Competition between genotypes of trees has been suggested (Tauer, 1975).

For commercial purposes, black cottonwood growth could be potentially developed in the form of 'very short' growth, 'short' growth, and 'long' growth. In each of these variations, the density of planting is less or more, and the diameter size (dbh) of

cut is either small or large (Verani et al., 2008). Spacing will affect the recovered biomass but mainly for early harvests; wood yield at 8 years proves to be better than 2-4 years cuts (Heilman and Peabody Jr., 1981). An alternative to complete harvest is the use of coppice culturing whereby trees are cut at particular heights from the growth and then allowed to regrow from sprouts, then to be reharvested at a later date (Debell, 1975; Harrington and DeBell, 1984; Debell, 1990). Depending on the frequency of harvest, initial planting for coppice systems could be as close as 1' by 1' or up to 6' by 6'. Cuttings cycles can vary from 2-8 years. Usually, increase in yield is obtained from small spacing after the first cut (~2 years), but later the yield is not as affected by spacing. For cycles of 2-5 years, up to 400 cubic feet of tree can be harvested which can equate to seven dry tons per acre per year. Some coppice harvesting will use four two-year cycles. With the support of herbicides and fertilizers, the first two-year cut can yield 13.4 fresh tons/hectare/year, and the recut some two years later of the coppice growth can yield 20.9 fresh tons/hectare/year (Heilman et al., 1972). Coppicing is associated, however, with some tree die off, and this may average ~10-40% with the tree kill accumulating per coppice harvest (Harrington and DeBell, 1984). Initial observations suggested that coppiced black cottonwood might grow better in mixed plantations with red alder in contrast to pure black cottonwood stands (DeBell and Radwan, 1979; Heilman and Stettler, 1985). The latter benefit appears to also depend on existing soil conditions.

The presence of natural black cottonwood stands adjacent to the presence of black cottonwood hybrids in British Columbia has the potential to confuse the observer in regards to whether one is truly dealing with the native species or a hybrid. Natural hybrids can also occur. The ability to determine the genetic origin of single or multiple

trees is therefore very important at times in understanding the relationship of tree and disease. In this regard, Beauseigle et al. (2010) have identified that molecular techniques which are based on sequencing of *Populus* genes can discriminate among species and hybrid parent genetics.

vi. Natural forest composites

P. trichocarpa can be associated in forests with many other tree species, especially those that are accepting and tolerant to the same moisture and soil requirements (DeBell, 1990). Willows (*Salix*) are the dominant trees associated with *P. trichocarpa*; examples include *S. lasiandra* (Pacific willow), *S. sessilifolia* (northwest willow), *S. fluviatilis* (river willow), and *S. scouleriana* (Scouler willow). In the mix of coastal forests, black cottonwood can be found in mixtures with *Alnus rubra* (red alder), *Pseudotsuga menziesii* (Douglas fir), *Tsuga heterophylla* (western hemlock), *Thuja plicata* (western redcedar), *Picea sitchensis* (Sitka spruce), *Abies grandis* (grand fir), *Acer macrophyllum* (bigleaf maple), *Fraxinus latifolia* (Oregon ash), *Crataegus douglasii* (black hawthorn), *Betula* spp.(birch), and *Prunus* spp. (cherry). In forests more inland, other associations include *Pinus monticola* (western white pine), *Pinus ponderosa* (ponderosa pine), *Abies concolor* (white fir), *Larix occidentalis* (western larch), *Abies lasiocarpa* (subalpine fir), *Picea glauca* (white spruce), *Picea engelmannii* (Engelmann spruce), and *Populus tremuloides* (quaking aspen). DeBell (1990) also details numerous shrub species and other herbaceous associations.

Smith (1957) has made some generalization that the species associated with black cottonwood are a reflection of the quality of the growth site. Therefore, very fertile land

will have black cottonwood associated with salmonberry, nettle, swordfern, lady fern, beaked hazel, and elder. Lesser quality soils will support black cottonwood with dogwood, honeysuckle, snowberry, and Nootka rose. If there is considerable flooding of the area, horsetails may be the dominant association.

vii. Hybrids of *Populus trichocarpa*

Both natural and artificial hybrids can be created for *P. trichocarpa*. Although early studies suggested that native *P. trichocarpa* was reasonably comparative in growth to common hybrids (Silen, 1947), it is now generally recognized that F1 hybrids with a *P. trichocarpa* parent are better growing than either parent (Heilman and Stettler, 1985a; Dickmann and Kuzovkina, 2008). The first trials of poplar hybridization in Canada appear to have dated back to the 1930s (Heimburger, 1936). Attempts in the latter studies with the purpose of crossing *P. trichocarpa* with *P. alba* were not successful, but researchers already recognized the benefit of using *P. trichocarpa* characteristics in crosses. The documented benefit of using *P. trichocarpa* in crosses was later more clearly established in Canada (Smith and Blom, 1966). Hybrid poplars which include *P. trichocarpa* traits are now commonly touted as being valuable for their fast growing ability and hence commercial value (Cisneros et al., 2000). There is considerable interest in developing hybrids which will provide maximal biomass for commercial purposes (Riemenschneider et al., 2001).

There are many naturally occurring *Populus* hybrids, but most hybrids have been derived from artificial hybridization with subsequent planting (Demerritt Jr., 1990; Slavov and Zhelev, 2010). Table 2 details commonly occurring natural hybrids. Of relevance to British Columbia, *P. balsamifera* naturally hybridizes with *P. trichocarpa* in

Table 2. Examples of naturally occurring hybrids among *Populus* in the world. Hybrids are by convention named with the female parent first and male parent second (from Demeritt Jr., 1990; Slavov and Zhelev, 2010).

Hybrid name	Parental poplars	Common name (if any)
P. x *tomentosa*	*P. alba* x *P. adenopoda*	
P. x *roulwauiana*	*P. alba* x *P. grandidentata*	
P. x *canescens*	*P. alba* x *P. tremula*	
P. x *heimburgeri*	*P. alba* x *P. tremuloides*	
P. x *brayshawii*	*P. angustifolia* x *P. balsamifera*	
P. x *acuminata*	*P. angustifolia* x *P. deltoides*	Lanceleaf Cottonwood
P. x *andrewsii*	*P. angustifolia* x *P. deltoides*	
P. x *hinckleyana*	*P. angustifolia* x *P. fremontii*	
P. x *sennii*	*P. angustifolia* x *P. tremuloides*	
P. x *jackii*	*P. balsamifera* x *P. deltoides*	Jackii Poplar
P. x *dutillyi*	*P. balsamifera* x *P. tremuloides*	
P. x *euroamericana*	*P. deltoides* x *P. nigra*	Euramerican Poplar
P. x *canadensis*	*P. deltoides* x *P. nigra*	Euramerican Poplar
P. x *bernardii*	*P. deltoides* x *P. tremuloides*	Bernard Poplar
P. x *generosa*	*P. deltoides* x ***P. trichocarpa***	Interamerican Poplar
P. x *interamericana*	***P. trichocarpa*** x *P. deltoides*	Interamerican Poplar
P. x *parryi*	*P. fremontii* x ***P. trichocarpa***	Parry Cottonwood
P. x *smithii*	*P. grandidentata* x *P. tremuloides*	
P. x *berolinensis*	*P. laurifolia* x *P. nigra*	Berlin or Russian Poplar
Unnamed	*P. deltoides* x *P. balsamifera* x *P. angustifolia* (natural trihybrid)	Unnamed

(Note: some literature citations designate *P.* x *generosa* as *P. deltoides* x ***P. trichocarpa*** or as ***P. trichocarpa*** x *P. deltoides* and so confusion may arise. See Stanton et al., 2010)

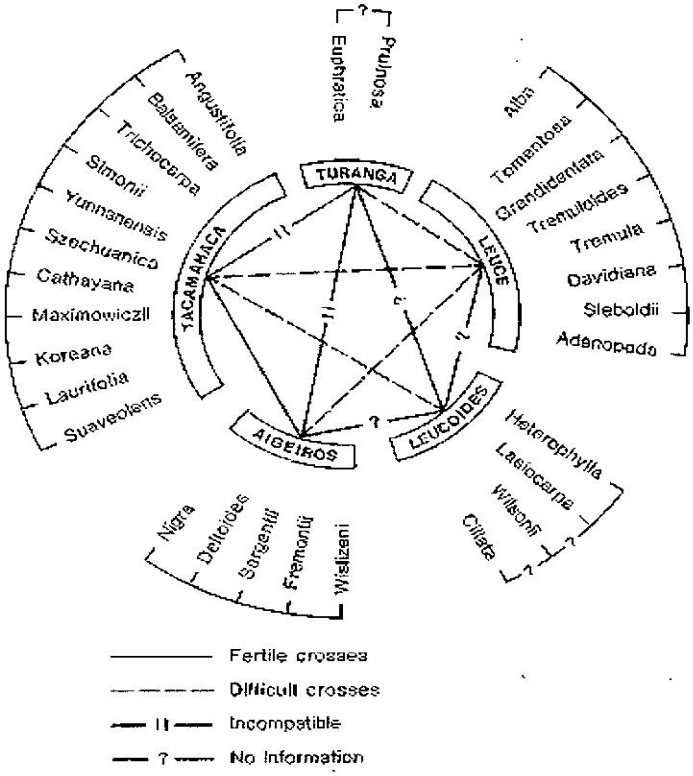
areas where the two trees overlap. In those areas where natural hybrids occur, the parent trees will be dominant to the hybrids. Hybrids generally grow well with the same soil conditions as the parents, and the phenology of the hybrids obviously is dependent on the parental characteristics that may be inherited. When reproducing hybrids, most will be achieved with growth from dormant cuttings which are harvested in January-February in the same year. They may be maintained in cold storage or frozen until planting, but the cutting should not be dried. Prior to planting, the cuttings are usually soaked in water for

one day. Good control of grass and weeds is important to survival in early growth. For example, grass competition can contribute to up to 50% mortality (Silen, 1947). It is better to cultivate the surrounding soil rather than apply chemical sprays. There is correlation to some extent of the early planting growth with later growth potential (Lo et al., 1995). Biomass for commercial use consists of oven dried leafless stems and branches. Hybrid biomass can be up to 50-140 tons/hectare after four years, and saw timber volume can be up to 145 cubic metres/hectare after 15 years (Dickmann and Kuzovina, 2008). Both fertilization and irrigation will promote the best biomass propagation. Hybrids may live up to 100 years.

Hybrids can be created from species within *Populus* sections but even between species from Sections. There are some restrictions, however, for the extent of such hybridization (Figure 13). Hybrid crosses may be better achieved with a designated female/male direction among Sections, e.g. using the *Aigeiros* Section female with the *Tacamahaca* Section male (Stanton et al., 2010). Hybrid crosses may also be better achieved with some direction in female/male parentage, e.g., *P. deltoides* x *P. trichocarpa* produces better than the reverse hybrid. Repetitive intercrossing of hybrids or back-crossing with parent trees may be attempted to improve on some of the genetic traits experimentally.

P. x *generosa* can have some 75-177% more stem volume by year two and up to 50% more stem volume by year four in comparison to *P. trichocarpa*. In the *P.* x *generosa* hybrid, *P. trichocarpa* genes confer larger leaf cells and *P. deltoides* contributes greater density of leaf cells, so that overall the hybrid has a larger leaf area than parents (Ridge et al., 1986). For the *P. trichocarpa* x *P. deltoides* hybrid, there can be up

Figure 13. Harmony of hybridization among *Populus* spp. from different sections (from Demerritt Jr., 1990).



to 1.5 times and 2.3 times the stem volume than the *P. trichocarpa* and *P. deltoides* parents respectively; the heights can be 1.1 and 1.3 times the parents respectively (Ceulemans et al., 1992). Hybrids have an intermediate number of sylleptic branches between the parents, the more of which are attributed to *P. trichocarpa*. *P. trichocarpa* parent is better than the hybrid for nitrogen and phosphorus utilization efficiency, thus suggesting the value of genes from the parent (Heilman and Stettler, 1986). The potential value for hybridization was shown by Bassman and Zwier (1991) who found that a

hybrid derived from Western Washington *P. trichocarpa* and drought resistant *P. deltoides* was optimal for growth in the drier interior compared to native *P. trichocarpa*.

F1 hybrids are mainly used for plantations since they generally outperform F2 hybrids (Stanton, 2009). In studies of *P. trichocarpa* x *P. deltoides* hybrids, researchers looked at comparisons of F1, F2, and backcrosses (*P. trichocarpa* x hybrid and hybrid x *P. trichocarpa*) (Stettler et al., 1988). Variation in trees was greatest for F2, intermediate for backcrosses, and least for the F1 hybrid. Coppice growth was better for the F1 hybrid. Parent *P. trichocarpa* performed better than F2 hybrids for four year growth volume.

viii. Uses of *Populus trichocarpa*

Traditional Uses:

P. trichocarpa has been used traditionally by First Nations people in British Columbia for many indications as cataloged by Pojar and MacKinnon (1994). These include:

- sweet inner bark and cambium could be eaten in late spring and early summer
- buds could be picked in spring, then boiled in deer fat to create a fragrant salve
- gum from buds were used in various medicinal preparations
- buds could be used as a poultice
- old leaves could be boiled for use in a medicinal bath
- wood used as a fuel for smoking fish
- antiseptic properties: leaves used on cuts and boiled bark used for sore throats
- inner bark used to reinforce other plant materials for spinning
- construction of small dugout canoes
- bud gum used for waterproofing of baskets and boxes
- soap and hairwash made from ashes
- bark strips used to make buckets
- roots used for ropes and fish traps
- bud gum used as a glue
- buds boiled in grease and mixed with pigment to make paint

P. trichocarpa contains salicin which is the natural predecessor to aspirin-like compounds and hence the true medicinal quality of the tree.

Practical non-commercial uses:

P. trichocarpa may be used in several settings if not only for their fast growth and large size. In these respects, the species may be used analogous to other *Populus* species or similar rapid growing large trees such as their hybrids (DeBell, 1990;Demeritt 1990). Such practical applications can include:

- shading
- landscaping
- bordering
- soil stability on hillsides
- windbreaks around housing, especially on farms and acreages
- windbreaks for reduction of soil erosion by winds
- studying the effects of air pollution
- firewood

There is much competition for trees to be used as ornamental plants, and in this regard, *P. trichocarpa* is not a common selection.

Commercial uses:

P. trichocarpa, when grown to sufficient size and desirable quality, may be used in many commercial functions. These functions are shared with many other *Populus* species and their hybrids. Commercial uses may draw from natural stands or from purposeful silviculture plantations. Two major commercial uses are in the production of wood and related structural media and in the production of paper (DeBell, 1990;Demerritt, 1990).

Wood Products:

The grain of *P. trichocarpa* is straight, fine, and even textured. The wood colour is light and is generally light weight. The wood is reasonably stable and tenacious for its weight, although its strength may be lesser than other woods. It has good nailing characteristics and does not split easily. It also peels well for veneer. The technologies are emerging such that products from smaller poplar, such as oriented strandboard, may rival traditional plywood made from high quality conifers. Wood engineering has been emerging considerably in the *Populus* area. Comparative aspects of poplar wood properties, processing, and utilization are detailed by Balatinecz et al. (2010). Given the uses as detailed below, it is possible to use all components of a log.

P. trichocarpa wood is generally of lower density than wood of other *Populus* spp., but its shrinkage potential is about the same. The density is similar to common softwoods (Table 3). Mechanical properties are relatively low compared to other hardwood and softwood sources, but the strength to density ratio is similar.(Table 3) Considerable variation, however, can be potentially achieved through hybrid production. The wood has high cellulose content (~80%) and low lignin content (~20%). The pH of poplar wood (~5.8-6.4) does not lead to metal corrosion nor to reactions with glue or coating products. Decay resistance is less than some other woods, which would suggest that preservative treatment of this wood might be required for some circumstances. Poplar wood machines well and does not require considerable energy for the same. The wood can be dried quickly if need be. High wood porosity allows poplar wood to bond easily with commercial adhesives.

Table 3. Physical and mechanical properties of mature wood from *P. trichocarpa* in relation to other common hardwoods and softwoods in North America (adapted from Balatinecz et al. 2010).

Physical properties

Species	Specific gravity	Shrinkage (green to OD) %			Side hardness
		radial	tangential	volumetric	
P. deltoides (eastern cottonwood)	0.37	3.9	9.2	13.9	1900
P. tremuloides (trembling aspen)	0.35	3.5	6.7	11.5	1870
P. trichocarpa	**0.31**	**3.6**	**8.6**	**12.4**	**1600**
Salix nigra (black willow)	0.36	3.3	8.7	13.9	-
Picea glauca (white spruce)	0.37	3.8	7.8	11.8	2100
Pinus banksiana (jack pine)	0.40	3.7	6.6	10.3	2500

Mechanical properties

Species	Flexural		Compression strength (MPa)		Shear strength (MPa)
	strength (MPa)	modulus (GPa)	parallel	perpendicular	parallel
P. deltoides (eastern cottonwood)	59	9.4	33.9	2.6	6.4
P. tremuloides (trembling aspen)	58	8.1	29.3	2.6	5.9
P. trichocarpa	**59**	**8.8**	**31.0**	**2.1**	**7.2**
Salix nigra (black willow)	54	7.0	28.3	3.0	8.6
Picea glauca (white spruce)	68	9.2	37.7	3.2	7.4
Pinus banksiana (jack pine)	68	9.3	39.0	4.0	8.1

Note: ‘parallel’ and ‘perpendicular’ refer to strength relative to the grain.

Wood products include potentially at least the following:

a) lumber: When bark-free logs are utilized, lumber recovery is approximately 45-50% with the remaining in chips (~40-50%) and in residual sawdust (~5%). Only ~15% of poplar lumber is in the high value top grades and ~60% in the low grades. Internal parts of furniture are becoming a use for poplar wood. Kiln drying techniques require improvement, but this is achievable and has been assessed at the University of British Columbia for hybrid *P. trichocarpa* as acquired from Harrison Mills plantations (Kang et al. 2007).

b) wood-based composites and panel products:

- veneer and plywood – requires high quality logs, but poplar is very well-suited for such peeling.
- fibreboard – these products include insulation board, hardboard, and medium density fibreboard. The materials used to construct these are fibres and fibre bundles that usually arise from residue of planer shaving, sawdust, and wood chips. The fibre is not as large as particles, flakes, and strands which are used for other purposes. The fibre is obtained by thermo-mechanical pulping.
- particle board – this product is made from particles and flakes as a composite, whereby the product is held together with adhesives. Poplar wood can be blended with other wood sources since it is dependent on available residue.
- oriented strandboard – this product is becoming more widely manufactured in B.C. and used world-wide. The product is highly engineered and can be produced from small-sized logs. Strands, large flakes, and wafers of wood are organized in a layered construction in which surface layers have strand orientation in one

direction and where inner layers have random stranding. As the availability of good peeling logs of the softwood variety become more limited in the plywood domain, a move to oriented strandboard becomes more appealing. Black cottonwood appears to produce inferior oriented strand board compared to aspen, but hybrids of this tree are improved and promising (Groover et al., 2010). Semple et al. (2007) have shown that fast grown large diameter wood of initial lower density from *P. trichocarpa* hybrid poplars in the Fraser Valley can be suited to produce this product.

- wood-cement products – various wood particles are bonded with Portland cement or similar. Such composites may be more resistant to weather elements, fire, and living pests such as insects and fungi. The wood component insures a lighter weight than would be achieved with concrete alone.
- wood-plastic products – various plastic polymers are mixed with small particle wood. Recycled plastic may be used in the process. The applications are ever increasing, and demand is fueled by the 'green' drive.

c) specialty wood products:

Pallets are a common use for poplar wood. Other possibilities include the need for light weight wood for tongue depressors, chopsticks, and other similar small products.

d) paper products:

The short fine fibers of *P. trichocarpa* facilitate the production of a pulp that is suitable for high-grade book and magazine papers. All commercial pulping methods can be applied to poplar. Poplar is used in: a) specialty papers – towlettes, napkins, tissue

paper, b) newsprint and similar paper when mixed with softwood pulp, and c) paper board for packaging, insulation, and ceiling tiles. Poplar hybrids are particularly useful in these regards.

New Sources for Energy:

Expedient growth of poplars in a plantation with minimal input provides for a biomass that can potentially replace fossil fuel. The future prospect for such energy depends considerably on cost, investment, taxation, and the long-term availability of fossil fuels. Energy production from poplar wood could conceivably occur by three main processes: a) thermal or direct combustion, b) biological degradation to form ethanol or methane, and c) physical degradation to create further products which can ultimately be converted into ethanol. Densification to fuel pellets provides a more efficient product than raw wood in the thermal use of poplar. Although corn use for ethanol production is receiving much attention, some project that cellulosic ethanol can produce up to 3-4 times the amount of ethanol per hectare in comparison.

Overall, the potential here is considerable given that production and use are effectively CO_2 neutral.

Renewed Means for Carbon Sequestration:

The sequestration of carbon dioxide from the atmosphere is taking on considerable interest from those who deem it necessary to slow climate change. The rapid growth of *P. trichocarpa* and other poplars and their large potential size offer some potential in this regard.

Perfume:

Extracts of buds can be used in perfumery and cosmetics.

Animal Feed Pellets:

Pellets for ruminant animals in capture may include poplar wood.

ix. Diseases of *Populus trichocarpa* in British Columbia

Table 4 highlights some of the more common pathogens and disturbances that hinder *P. trichocarpa* growth and survival in British Columbia. These have been beautifully and cogently detailed in the illustrated diagnostic manual of Callan (1998). On a more exhaustive basis, Table 5 details fungal pathogens that have been cited in B.C. for the host *P. trichocarpa*.

Non-infectious (abiotic) causes of tree damage result mainly from the environmental elements (wind, temperature, and atmosphere) or from environmental stressors (water and nutrient) either naturally arising or as propagated by human development (Marron et al.,2008). These insults can favour tree and/or foliage loss directly or indirectly. In regards to the latter, stressed trees and their acquired physical imperfections from such non-infectious causes may allow for pathogenic living organisms to invade more easily. For example, *S. musiva* may profit from bark anomalies to allow colonization and thereafter growth within the cambium. Conversely, the disease caused by living pathogens may facilitate tree destruction by environmental mechanisms. For example, *S. musiva* stem or branch canker may be so extensive so as to facilitate wind breakage at the diseased site.

P. trichocarpa is host to both vertebrate and arthropod pests (Smith and Bloom, 1966). Vertebrates are more likely to enhance damage when the tree is young. In both cases, damage to cambium may promote invasive fungi such as *S. musiva*.

Table 4. Classes of diseases of *Populus trichocarpa* in British Columbia (as per Callan, 1998).

Abiotic trauma

- temperature
- air pollution
- precipitation/hail
- moisture
- toxic/chemical
- pH excess
- wind
- direct trauma
- nutritional deficiency

Predators

- arthropods
  - *Harmandia* spp. – galls
  - *Cryptorhynchus lapathia* – poplar bore weevil
  - *Pemphigus* spp. – aphid leaf gall
  - *Dasineura* spp. – big bud midge
  - *Aceria parapopuli* – bud gall mite
  - *Schizoempodium mesophyllincola* – leaf bronzing mite
- vertebrates
  - ungulates
  - rodents
  - birds

Fungal diseases of leaves and shoot

linospora leaf blight – *Linospora tetraspora*
leaf rust – *Melampsora occidentalis*
taphrina leaf blister and curl – *Taphrina populina*
venturia leaf/shoot blight – *Venturia populina*
powder mildew – *Uncinula salicis*
marssonia leaf blight – *Marssonina populi*
septoria leaf blight – *Septoria musiva*
- *Septoria populicola*
minor leaf spot – *Phaeoramularia maculicola*

Fungal diseases of stem and branches

black stem – *Vassa sordida*, *Diaporthe eres, Colletotrichum gloeosporioides*
cytospora canker – *Vassa sordida*
neofabraea canker – *Neofabraea populi*
septoria canker – *Septoria musiva*
cryptosphaeria canker – *Cryptosphaeria lignyota*
branch galls – *Diplodia tumefaciens*

Wood decay fungi

butt rot – *Armillaria sinapina, Armillaria nabsnona, Ganoderma applanatum*
black gall – *Phellinus igniarius*
other basidiomycete decays – *Bjerkandera adusta*, *Coprinus* spp., *Daedaleopsis confragosa*, *Pholiota populnea*, *Pleurotus ostreatus*, *Radulodon americanus*, *Spongipellis delectans*, *Trametes versicolor*, *Trichaptum subchartaceum*

Table 5. Index of fungi associated with *Populus trichocarpa* in British Columbia (as per Callan, 1998).

Ascomycetes (fungi that develop their spores in sac-like structures called asci)

Amphisphaerella amphisphaerioides
Caliciopsis calicioides
Cryptodiaporthe salicella
Diaporthe eres
Hyaloscypha hyalina
Leucostoma nivea
Melanomma fuscidulum
Mycospharella populorum
Neofabraea populi
Peziza rapanda
Scutellina scutellata
Taphrina populi-salicis
Valsa sordida
Arachnopeziza sp.
Coccomyces sp.
Cucurbitaria staphula
Discina perlata
Hypoxylon multiforme
Linospora tetraspora
Mycosphaerella populicola
Mycosphaerella tassiana
Ophiostoma piliferum
Phaeocalicium populneum
Stictis radiata
Tympanis spermatiospora
Venturia populina
Ascocoryne sarcoides
Cryptosphaeria lignyota
Diatrype macounii
Eutypa maura
Hypoxylon vogesiacum
Lophodermium sp.
Mycosphaerella populifolia
Nectria inventa
Peziza emileia
Pleospora sp.
Taphrina populina
Uncinula adunca

Coelomycetes (fungi that produce conidia in pycnidiae)

Cyrptosporiopsis sp.
Cytosporina sp.
Discella microsperma
Phoma sp.
Septoria musiva
Cytospora chrysosperma
Dichomera sp.
Libertella sp.
Phomopsis oblonga
Sirodothis populnea
Cytospora nivea
Diplodia tumefaciens
Marssonia populi
Septoria populicola

Hyphomycetes (fungi that bear conidia freely on the mycelium)

Aegerita sp.
Aureobasidium pullulans
Cladosporium sessile
Epicoccum sp.
Phaeoramularia maculicola
Pollaccia elegans
Trichoderma sp.
Alternaria alternata
Cadophora sp.
Coryne sarcoides
Fusarium lateritium
Phialocephala bactrospora
Ramularia sp.
Verticillium tenereum
Aspergillus sp.
Cladosporium herbarum
Coryne dubia
Nodulisporium sp.
Phialophora sp.
Sporothrix sp.

Basiodiomycetes (fungi that bear their spores on outside fruiting bodies, like mushrooms)

Amphinema byssoides
Aporpium caryae
Bjerkandra adusta
Ceriporia viridans
Cerrena unicolor
Coprinus sp.
Antrodia serialis
Armillaria nabsnona
Botryohypochnus isabellinus
Ceriporiosis aneirina
Chondrostereum purpureum
Coriolopsis gallica
Antrodia xantha
Basidiodendron grandiniodes
Calocera cornea
Ceriporiosis pannocincta
Clitocybe truncicola
Corticium roseum

Table 5. continued.

Crepidotus fulvotomentosus	*Cristinia hellvetica*	*Cylindrobasidium leave*
Cyphellopsis anomala	*Dacrymyces deliquescens*	*Daedaleopsis confragosa*
Datronia mollis	*Datronia stereoides*	*Diplomitoporus lenis*
Exidia glandulosa	*Exidiopsis fuliginea*	*Flammulina velutipes*
Fomes fomentarius	*Fomitopsis pinicola*	*Ganoderma applanatum*
Gloeocystidiellum karstenii	*Gloeocystidiellum porosum*	*Gloeoporus dichrous*
Grandinia arguta	*Gymnophilus spectabilis*	*Hapalophilus nidulans*
Helicogloea lagerheimii	*Hericium coralloides*	*Heterochaete spinulosa*
Hyphoderma inusitata	*Hyphoderma mutatum*	*Hyphoderma sambuci*
Hyphoderma setigerum	*Hypochnicium analogum*	*Hypochnicium vellereum*
Hypsizygus tessallatus	*Hypsizygus ulmarius*	*Inocybe* sp.
Inonotus glomeratus	*Inonotus obliquus*	*Intextomyces contiguus*
Irpex lacteus	*Junghuhnia nitida*	*Kuehneromyces mutabilis*
Laeticorticium expallens	*Lentinellus ursinus*	*Lentinellus vulpinus*
Marasmius tremulae	*Melampsora occidentalis*	*Meruliopsis corium*
Oxyporus corticola	*Oxyporus similis*	*Panis rudis*
Peniophora aurantiaca	*Peniophora polygonia*	*Peniophora rufa*
Phanaerochaete carnosa	*Phanaerochaete sordida*	*Phanaerochaete tuberculata*
Phellinus ferreus	*Phellinus igniarius*	*Phellinus viticola*
Phlebia albida	*Phlebia radiata*	*Pholiota populnea*
Pleurotellus hypnophilus	*Pleurotis dryinus*	*Pleurotis ostreatus*
Pleurotis subareolatus	*Pluteus atricapillus*	*Polyporus badius*
Polyporus elegans	*Polyporus melanpus*	*Polyporus squamosus*
Postia caesia	*Protodontia oligacantha*	*Pseudoclitocybe cyathiformis*
Punctularia strigoso-zonata	*Radulon americanus*	*Schizophyllum commune*
Schizopora paradoxa	*Scytinostroma galactinum*	*Sistotrema brinkmannii*
Sistotrema raduloides	*Spongipellis delectans*	*Spongipellis spumeus*
Steccherinum ciliolatum	*Steccherinum fimbriatum*	*Steccherinum ochraceum*
Stereum ostrea	*Subulicystidium longisporum*	*Tomentella calcicola*
Tomentella coerula	*Tomentella ferruginea*	*Tomentella jaapii*
Tomentellastrum badium	*Trametes hirsuta*	*Trametes pubescens*
Trametes suaveolens	*Trametes versicolor*	*Trechispora mollusca*
Tremella mesenterica	*Trichaptum biforme*	*Trichaptum subchartaceum*
Tulasnella calospora	*Typhula* sp.	*Tyromyces galactinus*

On a quantitative scale, tree damage and loss is more likely to occur secondary to fungal diseases of trunk, stems, shoots, and leaves. Some diseases, while a nuisance, will have less effect year-to-year on tree productivity, e.g., minor leaf spot or powdery mildew. Across North America, the two pathogens associated with most effect on poplar are *Melampsora* spp. (leaf rust) and *Septoria* spp. (leaf blight and canker) (Bjorkman, 1964; Wang and Van Der Kamp, 1992; Royle and Ostry, 1995). Historically, *S. populicola* caused most diseases of black cottonwood in B.C. (leaf blight but not

cankers). This review paper will later detail the emergence of *S. musiva* causing both leaf blight and canker in this province. When *S. musiva* causes canker, the resulting damage can invite other secondary fungi to invade and further the pathology.

Wood decaying fungi can belong to the basidiomycetes class which morphologically appear as mushroom-like structures and gall (Thomas and Podmore, 1953). Quantitatively, such disease does not lead to dramatic biological mass loss, although the appearances on tree bark can be quite dramatic. Infection can occur among fallen trees, and there is a tendency to affect older trees.

For diagnostic features of these diseases, the reader is directed to the manual of Callan (1998). On a preliminary basis, some diseases can be confused for one another. These diseases may be non-selective or, on occasion, may affect native *P. trichocarpa* or related hybrids differently. Some pathogens will affect stool beds or newly planted cuttings.

x. Host immunity and disease resistance in *P. trichocarpa*

The understanding of fungal immunity and *P. trichocarpa* is not well established on a general basis, and even less is known about the specific *Septoria* spp. – *P. trichocarpa* interaction. It has been recently established, however, that there is considerable potential for genetic variation among *P. trichocarpa* provenances for British Columbia, and such opportunity for study has been made possible by forward-thinking geneticists at the University of British Columbia, Vancouver (Gilchrist et al., 2006). Equal to the task of understanding the genetic basis of variability among local provenances, heterogeneity among phenotypic traits, e.g., physiological attributes, are

also being explored (Gornall and Guy, 2007). These pioneering studies, along with the recent complete sequencing of the *P. trichocarpa* genome, hold considerable promise for elucidation of *P. trichocarpa* responses to *Septoria* spp.

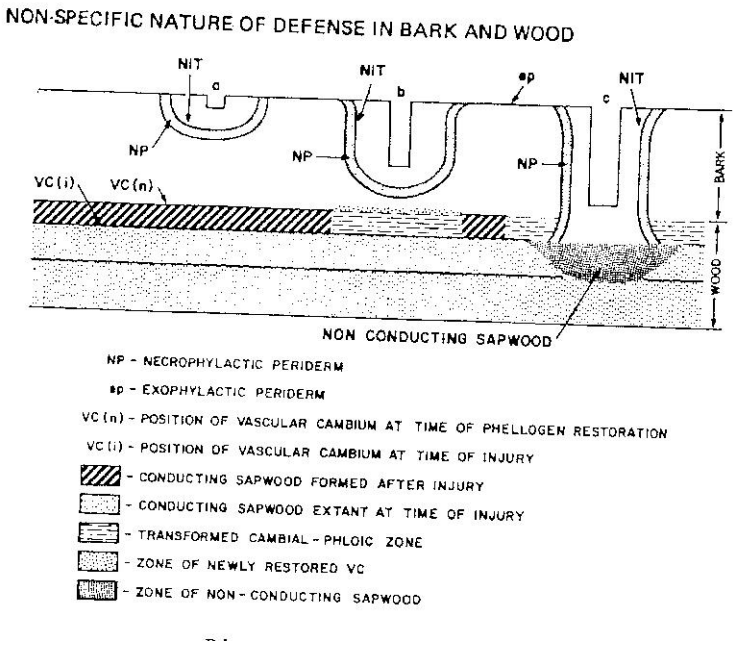
P. trichocarpa is exposed to innumerable living pests in nature, and yet relatively only a few of these cause damage among natural or plantation stands. On one hand, there may be a natural resistance based on the existing structure and function of these trees, e.g., the natural physical barrier of the outer bark. As it was historically recognized that particular *P. trichocarpa* hybrids were more resistant to many pathogens, it seemed that the genes for said resistance were commonly acquired from the exotic parent of the hybrid. Such resistance did not apparently co-evolve with exposure to the given pathogen. The latter is referred to as "exapted resistance", and it is not favoured by natural selection; it may have evolved for other functions. Contrasting the latter is the "adapted resistance" in which susceptibility develops between a fungal-tree combination where the context has allowed a co-evolution of both. That is, the co-presence over a long period of time has led to changes which allow the pathogen to evolve towards more propensity to cause disease or to escape host resistance strategies. One must recognize, however, that *Septoria* spp. multiply in generations exponentially greater than *P. trichocarpa* stands, and the potential for spontaneous mutation over this time is considerably advantageous for the fungi. Therefore, if *S. musiva* was more newly introduced into British Columbia, *P. trichocarpa* could be initially susceptible, all or none, or intermediately so, but the fungus would have considerable advantage to potentially change its invasiveness while it multiplies in such exponential numbers over many years in contrast to the longevity of a black cottonwood stand. If one looks solely at

the example of *Melampsora* rust in poplar hybrids, the simple fact that immature leaves have rust immunity compared to developed leaves suggests that some form of barrier or immunity must exist (Johnson and Kim, 2005). Others have indicated that resistance can be all or none or quantitative (Newcombe and Bradshaw, 1996; Newcombe, 1998). Regardless, the essential question is whether *P. trichocarpa* host responses can be modified to enhance resistance to *Septoria* spp. Mohamed et al. (2001) have attempted to turn on such resistance by overexpression of some resistance gene functions by genetic manipulation in *P. trichocarpa* parent hybrids; resistance to *S. musiva*, *S. populicola*, and other fungi was not achieved.

The impervious nature of tree bark serves as a first defence. *Septoria musiva* causing canker must initially enter through a wound or other breach of the outer impervious layer (Long et al., 1986). Physical barriers are innate to plants (and *P. trichocarpa*), and a non-specific reaction can be expected given the findings of non-specific reactions in other plant species. Mullick (1977), then of the Canadian Forestry Services in Victoria, detailed these non-specific events as per Figure 14. After penetrating the impervious layer, or phellem, and slightly below, there is an effort by the tree to restore the underlying cork cambium (phellogen). A "necrophylactic periderm" forms in which a layer of dying cells creates a thickening barrier. If the depth of the wound is furthered, then there is a process of vascular cambium restoration as well. Yet deeper injuries are also accompanied by the blockage of sap conduction within the wood. Biggs et al. (1984) demonstrate how this concept can be applied to fungal invasion (Figure 15). Zalasky (1964) proposed that such nonspecific host response could be demonstrated

between *P. trichocarpa* and *Macrophora tumefaciens*. Beyond such general resistance, which is yet to be specifically proven for *Septoria* spp. – *P. trichocarpa* interaction, it is

Figure 14. Diagrammatic views of the non-specific nature of defence during: (a) penetration of bark surface, (b) penetration of vascular cambium, and (c) penetration of sapwood. NIT = non-suberized impervious tissue/phellogen restoration (from Mullick, 1977).



likely that other signals for resistance should emerge. For example, "R (resistance) genes" may be signaled (Nimchuk et al., 2003). The latter would require either secondary local or distant signals to be activated, translocated, and responded to (Rathjen and Moffett, 2003; Thordal-Christensen, 2003). Figure 16 shows a model for the evolution of infection.

The presence of natural or recruited antimicrobial compounds, whether directly targeted or as metabolites, must also be a topic for future research in the *S. musiva – P. trichocarpa* model. *Populus* non-specific resistance analogous to the above is illustrated

Figure 15. Schematic diagrams of the tissues formed during phellogen regeneration in *Populus* bark following inoculation with fungus. 1 – movement of fungus through poorly lignified zone and away from the well lignified zone. 2 – impervious tissue forms in the inner region of the lignified zone. 3 – necrophylactic periderm (NP) forming internal to and next to the previously formed zone of impervious tissues (adapted from Biggs et al., 1984).

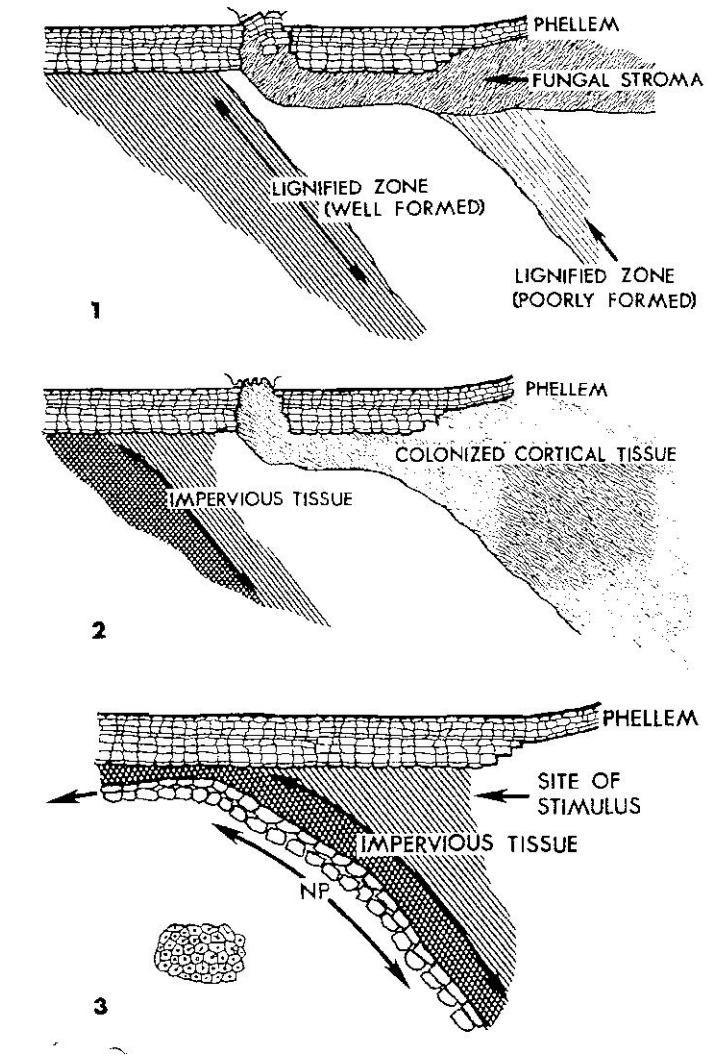
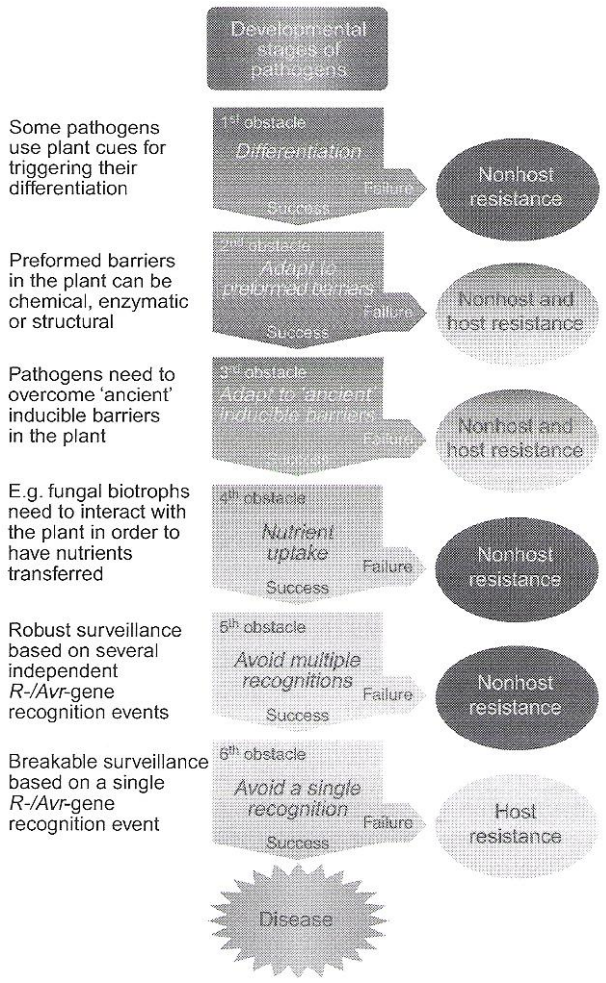


Figure 16. A model for the development of infection including both nonspecific and host specific resistance mechanisms (Thordal-Christensen, 2003).



in the example of healing cankers where lesion development is slowed and a lignified swelling results (Figure 17).

Table 6 outlines the recent published literature that otherwise might give some insight into the immunogenetics of *P. trichocarpa* and with some direct or indirect relevance to *Septoria* infection. The evolution of this work shows an initial understanding

Figure 17. Lignified swelling in response to *Septoria musiva* canker formation representing healing in a canker tolerant clone of *P. deltoides* X *P. maximowiczii* (from Feau et al., 2010, credited to Danielle Lamontagne, Point Platon, Quebec).

of genetic linkages for resistance and a progression towards working out the molecular bases for the same. Rapid progress in this regard is exemplified by the recent findings of Miranda et al. (2007) who have found transcriptional responses to flavonoid pathways evolving as *Melampsora medusae* causes leaf rust in *P. trichocarpa* x *P. deltoides* hybrids. In other tree systems, regulation of genes responsible for cell wall changes or antimicrobial proteins and metabolites (e.g., chitinases, trypsin inhibitors, polyphenol oxidase, terpenoids) has been recognized.

Table 6. Studies relating to the immunogenetics of *P. trichocarpa* and with some direct or indirect relevance to *Septoria* spp. infection.

Host	Findings	Implications	Reference
P. trichocarpa hybrids	no linear relation between leaf spot/canker susceptibility, but clone variability	potential for a genetic system for *Septoria* resistance	Palmer and Schipper (1979)
P. trichocarpa (British Columbia)	clones from dry regions less resistant than clones from coastal-humid regions for *Melampsora* leaf rust	a basis for immunogenetic variability	Wang and Van der Kamp (1992)
P. trichocarpa x *P. deltoides*	*S. populicola* leaf spot resistance linked to *P. deltoides* genes	potential for selective genetic breeding	Newcombe and Bradshaw (1996)
P. deltoides	*P. deltoides* resistance to *S. musiva* not linked to leaf resistance for *S. populicola*	potential specialized adaptive or exapted resistances for *Septoria* spp.	Newcombe and Bradshaw (1996)
P. trichocarpa	susceptible to *S. musiva* when grown in Eastern Canada	endogenous *P. trichocarpa* susceptibility to *S. musiva*	Newcombe (1998)
P. trichocarpa	two genes (Mmd1 and Mxc3) identified for rust resistance	fungal resistance determinants	Newcombe (2005)
P. trichocarpa hybrids (Vancouver Island)	hybrids with some fungal resistance had genes from exotic parent species	fungal exapted resistance	Newcombe (2005)
P. trichocarpa x *P. deltoides*	activation of cellular kinases responding to chitosan (fungal cell wall) and *S. musiva*	molecular host response	Hamel et al. (2005)
P. trichocarpa hybrids	*P. trichocarpa* imparts greater resistance to caterpillars	arthropod exapted resistance	Tomescu and Nef (2007)
P. trichocarpa x *P. deltoides*	defence genes activated variably in roots and leaves to physical and chemical stimuli	"root-shoot" defence system	Major and Constabel (2007)

There are other new and exciting developments taking place after the full sequencing of the *P. trichocarpa* genome. A genome-wide metabolic pathway database for *P. trichocarpa* is being established (Zhang et al., 2010). cDNA clones of genes responding to insect pathogens have been assessed (Ralph et al, 2008). NBS-resistance genes have been identified in *P. trichocarpa* which relate to triggers for cell-associated death in disease containment and also systemic acquired immunity (Kohler et al., 2008; DaQiang W et al., 2009).

III. *Septoria populicola*

i. Geographic distribution of *S. populicola*

S. populicola is mainly distributed in North America, although rare citation from southern Africa has been made (Royle and Ostry 1995; Sinclair and Lyon 2005). The fungus is particularly more common in the north central continent and pacific northwest and usually distributes with the presence of *Populus balsamifera* and *P. trichocarpa*. In several areas, there is overlap with geographic distribution of *S. musiva*. This fungus has the potential to follow areas that are planted with plantation poplar hybrids or imports.

ii. Spread of *S. populicola*

It is generally accepted that the sexual form of this fungus develops in overwintered leaves that have fallen the previous year. This probably begins in late fall. Such a phenomenon is apparently a common phase among other leaf disease-causing

ascomycetes fungi. The sexual phase of *S. populicola* matures with the formation of ascospore-containing pseudothecia. In early spring, the ascospores are released especially among fallen leaves when temperatures are warming and with moist weather. Within several weeks of the new leaves unfolding, the leaves may become infected through their stomata as airborne ascospores are released from the overwintered leaves; most of this infection will take place on lower branches which are more proximal to the ground level pseudothecia (Zalasky 1978). As the infection develops on leaves, pycnidia rich in conidia (asexual phase) are formed by 3-4 weeks. The conidia, when released and then dispersed by water and wind, may secondarily infect other leaves and stems, especially young shoots. More infections appear to occur when spring and early summer is associated with higher than average rainfall (see Figure 26 re: Spread of *S. musiva*).

The role of birds, bark predators, and insects in disease spread is not studied but is unlikely to be capable of spreading the larger distribution of fungi that is required for such widespread potential leaf disease. One cannot disregard the potential for importation to a site from other geographic regions via imported poplars or hybrids.

iii. Diseases of *S. populicola*

Leaf spot:

Leaf spot of *P. trichocarpa* with *S. populicola* is common British Columbia (Beauseigle et al., 2010). After the infective phase of the fungus enters the leaf via stomata, a necrotic spot evolves (Thompson, 1941). The spots may be initially yellow to light brown, but thereafter progress to dark brown and jet black (Figures 18-21) (Callan BE, 1998). The leaf spots may have well-defined margins and will often form in parallel to the long axis of the leaf. As the spots enlarge, they are usually round, but the fungal

mycelium growth can be delimited by vascular bundles of leaf vein and midrib (Thompson, 1941; Zalasky 1978). Spots vary from 1-15mm. in width, although usually 2-5 mm., and eventually the necrotic leaf blotches may coalesce and the entire leaf can be necrotic. The centre of the spot may have a whitish or slightly yellow appearance. At the centre of these spots, one may visualize fungal pycnidia under microscopic examination (Figures 22 and 23). Leaf spotting may be isolated, or under the right conditions and susceptibility, the spotting may appear as widespread chlorosis or necrosis on most leaves (Newcombe et al. 1995). Over the growing season, there is an apparent upwards migration of involved leaves throughout the tree (Thompson, 1941). Premature defoliation may lead to significant reduction in photosynthesis.

In contrast to *S. musiva*, *S. populicola* does not have as wide a spectrum of poplars that it may infect (Thompson, 1941). Nevertheless, *S. populicola* has been shown to variably cause leaf spot among *P. balsamifera* provenances in Manitoba under greenhouse experimentation (Zalasky, 1978). The latter work also found leaf spot produced among eastern cottonwood (*P. deltoides*) seedlings. Beauseigle et al. (2010) found leaf spot in both *P. deltoides* x *P. trichocarpa* and *P. maximowiczii* x *P. trichocarpa* hybrids in the Fraser Valley.

The detection of infection can be accomplished with direct culture of the leaf spot or with the application of molecular techniques which detect *S. populicola* DNA in the leaf sample (Beauseigle et al., 2010).

Canker:

S. populicola canker is rare and has been noted mainly under experimental

Figure 18. Extensive *Septoria* leaf spots on a hybrid poplar (*P. trichocarpa* X *P. maximowiczii*) (from Callan, 1998).

Figure 19. Mild *Septoria* leaf spot on a hybrid poplar (*P. trichocarpa* X *P. deltoides*) (from Callan, 1998).

Figure 20. Coalescing leaf spots due to *Septoria* on a poplar hybrid (*P. trichocarpa* X *P. deltoides*) (from Callan, 1998).

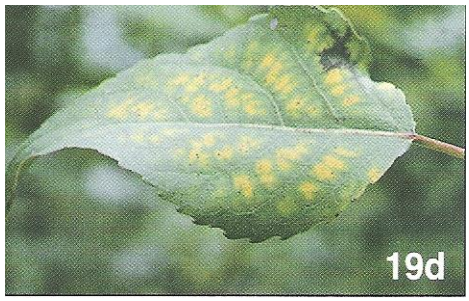


Figure 21. Early chlorosis which precedes the dark leaf spot due to *Septoria* on a poplar hybrid (*P. trichocarpa* X *P. deltoides*) (from Callan, 1998).

Figure 22. Pycnidia of *S. populicola* on a leaf of *P. trichocarpa* as seen in lower power microscopy. (from Callan, 1998).

Figure 23. Pycnidia of *S. populicola* on a leaf of *P. trichocarpa* as seen in lower power microscopy. Microscopy power is slightly higher than Figure 22 above (from Callan, 1998).

conditions. Cankers due to *S. populicola* are not documented in the wild on *P. trichocarpa* in British Columbia. Cankers secondary to this fungus can, however, be induced under experimental conditions (Zalasky, 1978). In the latter work, the authors caused canker in greenhouse conditions on *P. balsamifera* after aerosol inoculation. The appearance of canker associated with *S. populicola* mimics that of *S. musiva* in young seedlings/plantations (see below under *S. musiva*). Such cankers varied in frequency among *P. balsamifera* provenances from Manitoba.

iv. Treatment and prevention

Once leaf infection has taken place, it is conceivable that treatment of active disease may benefit the tree. Such an approach, however, has not gained interest, even despite the prospect of aerial spraying. An environmentally safe and economically feasible version of such a modality may yet be found. In the meantime, prevention rather than treatment has been the focus of scientific interest.

Silviculture practice in plantations has garnered most attention. Given the overwintering of fungus on fallen leaves, it may be of some value to remove leaf debris in the late fall. The use of resistant poplar clones and the assurance of disease-free stock may have merit. The early harvest and replacement of diseased clones could also be of assistance. Silviculturists should choose locations which are optimal for growth and yet less likely to bear the hazard for tree stress. Quarantine for tree imports must also be considered.

The enhancement of tree resistance has been forefront for scientists and foresters alike. Whether through natural selection, hybrid formation, or genetic or other biotechnical manipulation, a more fit poplar may be found.

Carbon (1972) examined the use of fungicides. In experimental plots of *Populus*, repetitive application of benomyl and thiophanate methyl derivatives proved to mitigate leaf spot formation due to *Septoria* spp. (no specific detail as to species singular or multiple prevented). Most regard this approach as one of high cost.

IV. *Septoria musiva*

i. General attributes

Septoria spp. are fungal pathogens of plants. They include over 1000 individual species. Although this review is focally concerned with disease among black cottonwood, *Septoria* spp. are notorious generally as leaf spot pathogens among foliage of vegetables, field crops, and forage, e.g., *S. tritici* affecting wheat. As fungi, the basic characteristics of *Septoria* spp. are eukaryotic. This distinction allows for a more complex genetic code than the more simplistic versions seen in bacteria and viruses. Such a complexity allows for potential adaptive infection and survival mechanisms and could feasibly provide for strategies in which the fungus could countervail man-made alterations in host plants including black cottonwood and its derivatives (e.g., crosses). The more complex genetic code also potentially complicates the use of antifungal agents since there is a more narrow window for toxicity between eukaryotes such as fungi and plants which could have cellular machinery mechanisms in common. Being a fungus, the generation times

for reproduction are short, and recombination/mutation events could possibly change the pathogen in a shorter period of time than may be seen among higher orders of eukaryotes.

Among fungi, *Septoria* spp. belong to the *Ascomycetes* phylum (division). This large group of fungi is defined in part by the sexual reproductive phase ("perfect state") which has a sac or "ascus" that contains "ascopores" from the sexual replicative process. This perfect state is called *Mycosphaerella*, and in particular, *Mycosphaerella populorum* for *S. musiva* and *Mycosphaerella populicola* for *S. populicola*. The *Septoria* (asexual) state produces spores in an arrangement called conidia (Bier,1939). These conidia are somewhat characteristic when seen under the microscope, and their size facilitates the differentiation of *S. musiva* from *S. populicola* (Figure 24) (Waterman, 1946). It is believed that the ascospore phase develops on dead leaves that are ground-laden after fall and that the conidial phase develops on new leaves directly (Thompson, 1941; Waterman, 1946). Regardless of stage, spores from either ascospores or conidia are able to aerosolize, thus providing a mechanism for spread to the same or other plants.

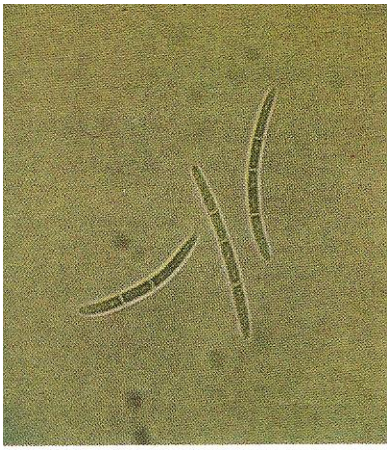


Figure 24. Conidia of *S. musiva* as seen by microscopy (from Palmer MA, et al., 1980).

Up to 10 *Septoria* spp. have been reported as being isolated from cottonwood sources, but in North America (and Argentina), *S. musiva* and *S. populicola* are associated with significant disease (*S. populi* in Europe, Asia, and occasionally Argentina). Beauseigle et al. (2010) have also recently suggested that a species identical or similar to *Mycosphaerella punctiformis* can be found rarely found in the Fraser Valley. *S. musiva* has now been identified across North America from coast to coast and as far north as Alaska, although it is particularly common in eastern and central United States/Canada.

ii. Geographic distribution

In contrast to *S. populicola*, this fungus is more common in east and central North America, especially Canada and the United States (Royle and Ostry, 1995; Sinclair and Lyon, 2005). It can be found as far north as Alaska. In Canada, the fungus can be found coast to coast. In British Columbia, it has been found in Fraser Valley plantations (Callan et al., 2007). New data from Beauseigle et al. (2010) suggest that the fungus may have spread to natural populations of black cottonwood from the latter plantations, although the latter isolations are rare. The fungus is absent in Europe, has been found in Argentina, and has been rarely cited in Asia. This fungus has the potential to follow areas that are planted with plantation poplar hybrids or imports.

iii. *S. musiva* in British Columbia

Newcombe et al. (1995) detail longstanding experience in which it is said that native *P. trichocarpa* does not have endemic *S. musiva* infection, whether in leaf spot or canker forms. Such observations relate to the Pacific Northwest including British

Columbia and as reinforced by those experienced in this province. The notion of endemicity, however, changed with the observations of Callan et al. (2007) when they found, and genetically confirmed, the presence of *S. musiva* in hybrid poplar (*P. trichocarpa* x *P. maximowiczii*) canker in a Fraser Valley plantation. Further observation of the latter continues to suggest that *S. musiva* is endemic in that small geographic region where Callan et al. made their findings. The latter, however, did not initially include native *P. trichocarpa*.

Table 1 of the Callan et al. paper, however, raises some interesting prospects. Apart from their findings of *S. musiva* in the Fraser Valley hybrids during 2006, the Table also cites *S. musiva* detection from leaf spot on five other occasions almost two decades earlier during 1988-1991. One of the citations DAVFP 23833 relates to microscopic observation of leaf spot from specimen obtained of *P. tremuloides* at Broman Lake (northwest of Prince George). Such observation depended on the finding of compatibly-sized conidia. Other microscopic examination found putative *S. musiva* (DAVFP 24241) on leaf spot from a Burns Lake plantation (northwest of Prince George) of *P. trichocarpa* and the same from *P. trichocarpa* (DAVFP 24242) in Rosswood (coastal B.C. due west of Prince George). Perhaps most important among these citations was the finding of *S. musiva* (DAVFP 24201 and DAVFP 24202) microscopically on poplar hybrids at Harrison Mills in the former Scott Paper nursery. Such an observation would date *S. musiva* presence some two decades in the least. Although of considerable interest, the importance of these findings is dampered by the limited technology of the time, that being dependence on microscopic examination. The conidia of *S. musiva* are smaller than *S. populicola*, and it is not clear that the sightings were either *S. musiva* or

possibly immature *S. populicola* conidia of similar and thus mimicking size. Callan et al. (2007) found *S. musiva* in 2006 but provided convincing evidence with both the culture of the fungus and with the use of genetic detection. Leaf spot samples from earlier herbarium filings are not available for molecular detection. Details of the herbarium are found at the Pacific Forestry Centre web-site (see below).

The finding of *S. musiva* on either leaf spot or canker elsewhere among native *P. trichocarpa* would possibly be of significant value in determining degree of spread and origin. Keiran (2008) has indicated recently how important the spread of *S. musiva* could be for both commercial tree stands or the natural forests.

The findings of Beauseigle et al. (2010) now add a new dimension. The latter authors again found *S. musiva* among trees in the Fraser Valley plantations including both *P. trichocarpa* and its hybrids. Rare findings of *S. musiva* in native black cottonwood outside of these plantations were also detailed. The latter raises the prospect of continued spread in this context. Of interest, the findings of *S. musiva* did not appear to be diffuse among the leaves of one tree, and thus the infections were either considered to be rare or of a mixed nature in the same tree and stand. Several identifications were also made in plantation trees that represented different *P. trichocarpa* provenances.

Other references to S. musiva and British Columbia:

Apart from the Callan et al. (2007) and Beauseigle et al. (2010) references as detailed above, Bier (1961) indicated that *Septoria* canker-like disease was not seen in the Vancouver area, but then suggested that cuttings of *P. trichocarpa* and *P. tremuloides* that were harvested from poplars in the University of British Columbia area were susceptible to *S. musiva* when subjected to drying and thus moisture reduction. Bier

(1939) also had earlier indicated the lack of natural *S. musiva* on the west coast of Canada, but recognized that artificial inoculation of black cottonwood leaves could experimentally induce leaf spot.

iv. Diversity in *S. musiva*

Phenotypic:

In an examination of *S. musiva* isolates from Ontario and the United States, there was an apparent similarity in such attributes as culture morphology, temperature requirements, and virulence as assessed by artificial inoculation-canker production in hybrids (*P. deltoides* X *P. trichocarpa*) (Spielman et al., 1986). Krupinsky (1989) assessed for phenotypic variation in 'aggressiveness' using a disease rating system for leaves in greenhouse hybrids. There were apparent differences between local isolates and other regional isolates. Mottet et al. (1991) found differences in aggressiveness of four *S. musiva* isolates during inoculation studies. Although there was a general trend for such variation among these four isolates for various poplarss, the authors also found at times that aggressiveness ranking of these isolates in one species or hybrid of tree was different than for others. The latter studies were also important in that they confirmed what other researchers had suggested previously in regards to the differential susceptibility of poplars. Generalizing, the Ageiros Section tended to be more resistant than poplars of the Tamacahaca Section. Hybrids between these Sections gave intermediate resistance. Ward and Ostry (1994) found variability in growth rate, color, morphology, and sporulation. The same authors later showed variation among *S. musiva* isolates for leaf-disc and stem infection assays among hybrids, although there were some findings suggestive of a dissociation between propensity/intensity of disease in leaves versus cankers (Ward and

Ostry, 2005). These findings gave rise to the belief that *S. musiva* variation might allow for differences in disease causation (Feau et al., 2010). Studies which have examined the protease production by *S. musiva* have been undertaken (Sillick et al., 1989).

LeBoldus et al. (2007, 2008, 2009) have provided data which must alter the focus of pathogen-host interaction. Firstly, they found disease severity to be similar among *S. musiva* from either Alberta or Quebec (LeBoldus et al., 2009). Examining hybrids in greenhouse conditions, there were no apparent differences for isolates based on geographic source, host tree source, or disease symptoms (LeBoldus et al., 2008). The latter included no difference among isolates whether acquired from hybrid or native trees. In similar studies, it appeared that variability was more likely linked to poplar genotype rather than fungal isolates (LeBoldus et al., 2007). The hybrids used in the latter three studies, however, did not have any *P. trichocarpa* parentage.

Genetics:

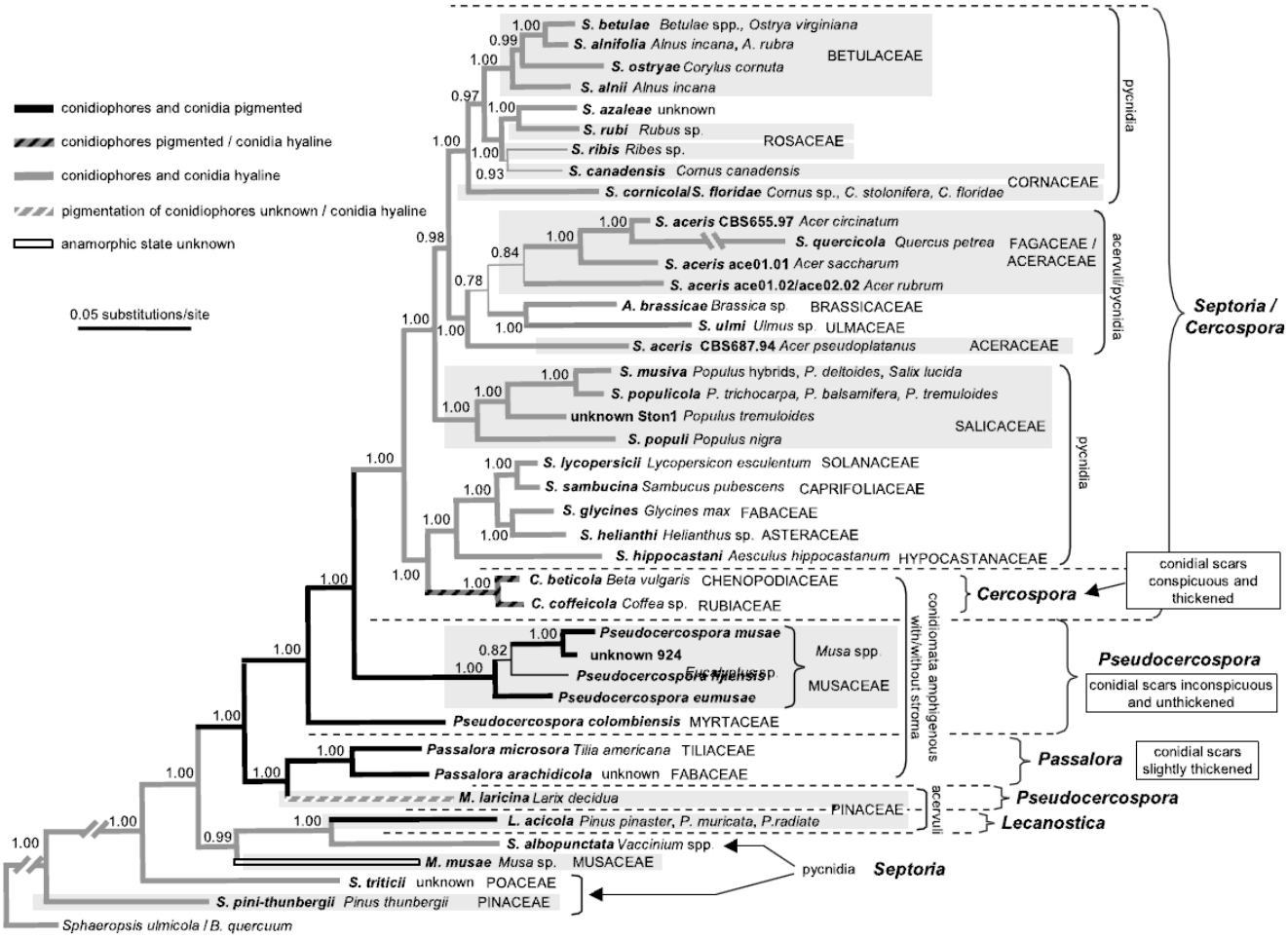
Given the existence of both sexual and asexual phases of *S. musiva* (*Mycosphaerella*), one might inherently presume the potential for considerable genetic variation (Feau et al., 2010). Nevertheless, it is generally not understood what role sexual and asexual phases have in genetic changes, i.e., regarding genetic drift and shift. In some areas of the *S. musiva* genome, there must be very conserved and slowly changing genes; such may be exploited for developing consistent genetic-based DNA detection/identification techniques (Feau et al., 2005b; Weiland and Stanosz, 2006; Callan et al., 2007). Beauseigle et al. (2010) indicate that both culture and molecular techniques can be used to find the pathogen in diseased plant tissue. Ward and Ostry (1994) used random amplification of DNA to generate DNA fingerprints and found

considerable variability (Ward and Ostry, 1994). Their later publications (Ward and Ostry, 2005) confirmed the latter, although acknowledging that there was overall considerable relatedness. The DNA fingerprint types were not related to host tree source, disease resistance, or geographic location. The collective findings led them to project that it was not incumbent upon *S. musiva* to adapt among different geographies.

In an analysis of randomly amplified polymorphic DNA fingerprinting of north-central and north-eastern North American isolates, Feau et al. (2005a) found moderate to high genetic differentiation but nevertheless could show a significant correlation between genotypes and geographic region where isolated. Leaf-canker paired isolates could be homogeneous, but different types could be identified on the same tree in different leaves. There was little difference among isolates from *P. deltoides* versus hybrid poplars. Other genetic studies supported the above data (Feau et al., 2006b). LeBoldus et al. (2009) have provided evidence of genetic similarity and dissimilarity for isolates from Alberta and Quebec. There is a correlation of differences among regions when one examines various gene products as markers for genetic variation (Spielman and Hubbes, 1984).

Examing *Septoria* spp. as a whole, it is clear that *S. musiva*, *S. populicola*, and *S. populi* are closely related, but *S. musiva* and *S. populicola* are most closely related (Figure 25) (Feau et al., 2006a). Further research has speculated on the potential for horizontal gene transfer among *Mycosphaerella* spp. (Feau et al., 2007a). The similarity of these species, however, will be of value where lessons from one may relate to another.

Figure 25. Tree diagram showing the relatedness of *Septoria* spp. to one another as assessed by a combination of genetic technologies in which gene structures are compared. The clustering of *S. musiva*, *S. populicola*, and *S. populi* is consistent with the similarity of the diseases that they cause in poplars. Note the clustering of these three species as compared to other fungi (from Feau et al., 2006a).



v. Spread of *S. musiva*

The spread of *S. musiva* is believed to occur as detailed for *S. populicola* (above) (Waterman, 1954b; Jones and Benson, 2001; Feau et al., 2010). In this regard, such spread has been more carefully studied by Luley and McNabb (1989 and 1991) and Ostry (1987). Many early and appropriate descriptions were also provided by Bier (1939). From the latter studies, it has been proposed that spread and secondary infection can occur for

3-4 months after initiation. The peak of ascospore release from pseudothecia correlates with peak leaf disease and stem/branch canker. Peak ascospore release occurs after rainfall and especially during daylight hours. Cankers only form on trees which bear leaf spots (Waterman, 1946). Although not absolutely confirmed, it is postulated that conidia from overwintered pycnidia in cankers might also cause secondary infections (Waterman, 1954b). Figure 26 illustrates the different infectious forms.

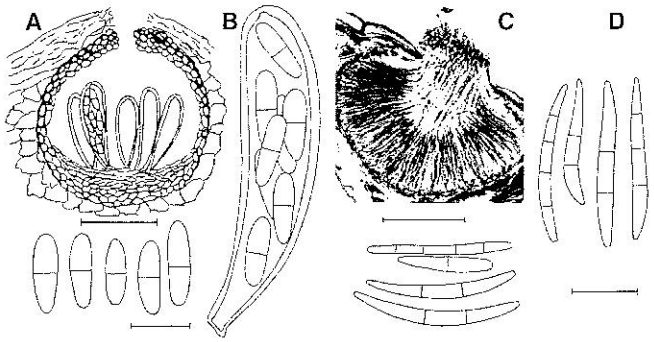


Figure 11. Mycosphaerella populorum. A. Section of pseudothecium. B. Ascus and ascospores. C. Section of pycnidium. D. Conidia. Scale bars = 50 µm for fruit bodies, 10 µm for other structures. A, B, D adapted from reference 3670 by permission of CAB International; C from reference 4017 by permission of the American Phytopathological Society.

Figure 26. Forms of *S. musiva* (*Mycosphaerella populorum*) that may exist in nature and which form as vehicles for spread in nature (from Sinclair and Lyon, 2005).

As for *S. populicola,* transfer from other living entities (e.g., birds) is not reported, but introduction from geographic areas via cuttings and seedlings, whether native or hybrid, must always be considered. Hildebrand et al. (2010) raise another very important potential source for *Septoria* spp. They have found that blueberry fields can be infested by leaf spot and stem canker due to *Septoria* spp. The latter species are not *S. musiva* (Alfieri, 1991), but nevertheless such findings raise some academic interest into whether *S. musiva* from poplars could cross over to other flora. In the Lower Fraser Valley, the

proximity of widespread blueberry cultivation and poplar tree farms poses risk for transfer in either direction.

vi. Diseases of *S. musiva*

Leaf spot:

S. musiva causes leaf spot similar to *S. populicola* (see above), but spots generally are larger (Thompson, 1941; Waterman, 1946; Cellerino,1999; Feau et al., 2010; Beauseigle et al., 2010). The pattern of leaf spot progression and subsequent defoliation is also similar (Cooper and Filer, 1976; Ostry and McNabb, 1985). Mature trees which have low branches that are far from the ground may be relatively spared for leaf spot (Waterman, 1954b). Specific photos of *S. musiva*-related leaf spot are published (Figures 27-33) (Palmer et al., 1980; Ostry et al., 1989; Sinclair and Lyon, 2005). Trees that bear some degree of fungal resistance may show smaller spots generally. Bier (1939) provided some of the earliest descriptions of leaf spot among poplars in Canada, and Thompson (1941) shortly followed by more extensive descriptions in the U.S. There are many species of poplars which are susceptible to *S. musiva* leaf spot.

The exact mechanisms of leaf spot production for *S. musiva* among poplars are not known, although some work has been already done in the area. For example, one report attributed the fungal production of ethylene and carbon monoxide as virulence factors for premature defoliation (Brown-Skrobot and Brown, 1984). Klepzig et al. (1997) have indicated that arthropod injury of leaves could enhance the potential for *S. musiva* to cause foliar disease among hybrid poplars.

Figure 27. Comparative assessment of poplars with and without *S. musiva* infection. The poplar on the left is significantly defoliated whereas the poplar on the right shows good resistance (from Ostry et al., 1989).

Figure 28. Young tree shows considerable leaf spotting due to *S. musiva* (from Ostry et al., 1989).

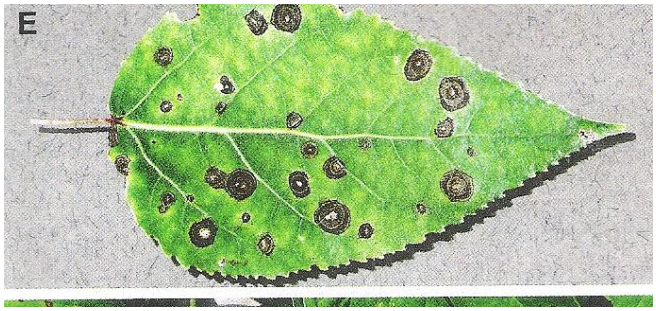
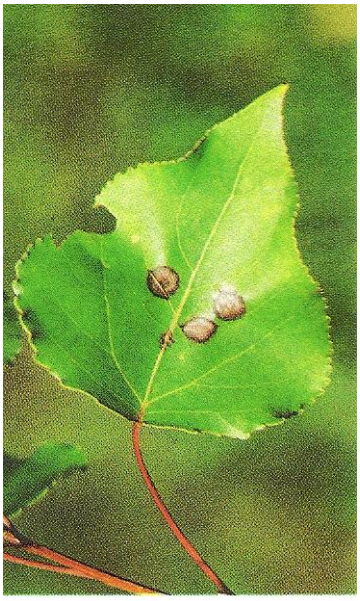


Figure 29. Example of leaf spotting on hybrid poplar due to *S. musiva*. Spot centres are grey-white (from Sinclair and Lyon, 2005).

Figure 30. Example of *S. musiva* leaf spotting showing larger sized spots. Spot centres are brown with bulls-eye focus (from Palmer et al., 1980).



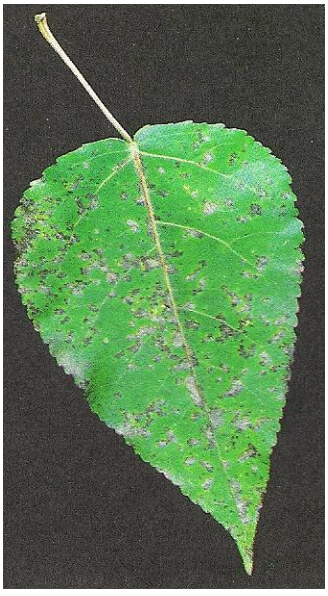
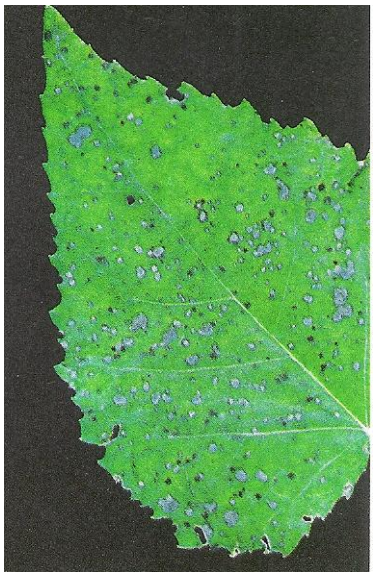


Figure 31. Example of *S. musiva* leaf spotting showing small spots that may coalesce (from Palmer et al., 1980).

Figure 32. Example of *S. musiva* leaf spotting showing small silvery spots (from Palmer et al., 1980).



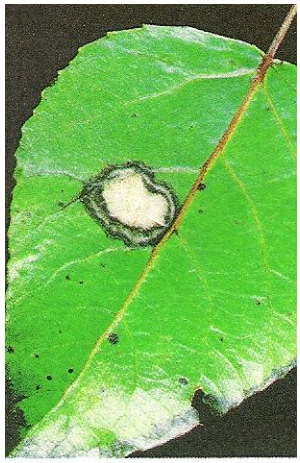


Figure 33. Example of *S. musiva* leaf spotting showing a large irregular spot and some marginal smaller spotting (from Palmer et al., 1980).

Canker:

S. musiva can induce stem or branch/shoot cankers in susceptible hosts (Figures 34-42) (Bier, 1939; Waterman, 1946; Waterman, 1954b; Zalasky, 1978; Mottet et al., 1991; Cellerino,1999; Jones and Benson, 2001; Feau et al., 2010; Beauseigle et al., 2010). The cankers are usually on the latter areas of poplar some 1-1.5 meters from ground level (Waterman, 1954b; Sinclair and Lyon, 2005), and it may be that high humidity close to the ground favours fungal growth and canker development. Multiple branches can be affected alone or in addition to the stem of the same tree. In coppiced plantations, new shoots are commonly affected in very susceptible poplar clones (Figure 36) (Ostry et al., 1989). Cankers are more likely to form on each year's new young shoots and lesser so in the two-year old branches (Waterman, 1954b). In nurseries and plantations, the young plants are more likely to succumb to infection in contrast to old trees (Long et al., 1986).

Cankers may, but do not usually, girdle the stem (Waterman, 1946; Waterman, 1954b). Stems with less than ¾ inch diameter may however be girdled in one growth season. The rapid growth of the tree may act as a defence mechanism to prevent the infection from girdling the stem. Secondary invasion of the initial *Septoria* canker provides an opportunity for a new wave of other fungi to attack (e.g., *Cytospora*, *Dothichiza*, *Fusarium*, *Phomopsis*) (Figure 37) which then may lead to complete girdling and possible stem breakage (Figure 38) (Waterman, 1946; Palmer et al., 1980; Ostry and McNabb, 1985). If breakage does not occur, facilitated by winds and other environmental conditions, the stem and branches and leaves distal to the canker can be compromised in

growth and degree of foliage if not altogether (Figure 39). Certainly an impediment of growth can arise. Abrahamson et al. (1990), in examining the impact of various diseases

Figure 34. *S. musiva* canker on the stem of a young poplar (from Ostry et al., 1989).

Figure 35. *S. musiva* canker on the stem of an older poplar (from Palmer et al., 1980).

Figure 36. Cankers on stump sprouts as would be seen in new growth from coppice plantation (from Ostry et al., 1989).

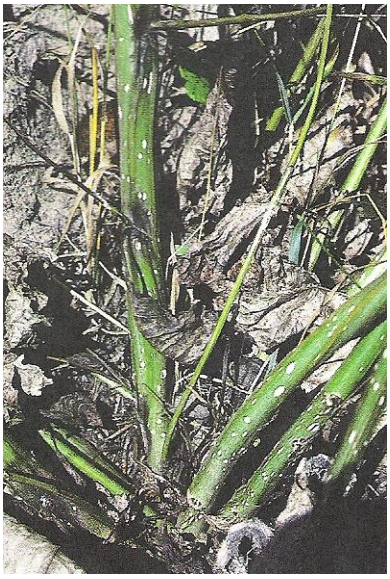


Figure 37. An old mature canker with some tree resistance likely secondarily infected with other fungi in addition to *S. musiva* (from Ostry et al., 1989).

Figure 38. *S. musiva* canker on a branch showing complete girdling effect (from Ostry et al., 1989).

Figure 39. Dieback in a poplar. The canker is lower in the stem and the dieback effect relates to distal growth towards the tree top (from Ostry et al., 1989).

among hybrid poplars, has suggested that canker severity is the major factor influencing tree growth potential.

Infections begin at bark wounds, although wounds are not always necessary (Waterman, 1946; Waterman, 1954b; Zalasky, 1978; Long et al., 1986; Feau et al., 2010). Lenticels, stipules, and leaf bases are foci for infection as well. Initially the canker will appear as a depressed area in the bark, perhaps even taking on the appearance of a 'water-soaked' region (Figure 40). The edge of the canker can be raised and discoloured black or slightly orange (Figure 41). Old cankers may appear ashen white. Pycnidia may be found at the centre of the canker but usually only in young cankers. The canker can elongate up and down the stem or branch/shoot. On side shoots, the canker can extend to the stem. Where the tree has developed some resistance, and especially to be seen on resistant clones, the canker formation may be halted and side areas of lignification can occur (Figure 42) (Waterman, 1954b). The latter represents tree healing. Although the distal growth can be impacted, a major degree of restoration can be achieved. One cannot isolate the fungus from the latter stage of canker (Waterman, 1954b).

Under experimental conditions, infection is easily produced with the initial wounding of the bark along with artificial inoculation. Trees that are atomized with conidia by aerosol only do not develop canker readily, but it is nevertheless possible (Zalasky, 1978; Long et al., 1986). *P. trichocarpa* canker due to *S. musiva* has been experimentally achieved in British Columbia with clones under experimentation at the University of British Columbia, Vancouver (Bier, 1961). Hybrids can be very susceptible (Ostry, 1987; Mottet et al., 1991).

Figure 40. Water-soaked appearance of bark disease in a poplar which is proximal to *S. musiva* canker (from Ostry et al., 1989).

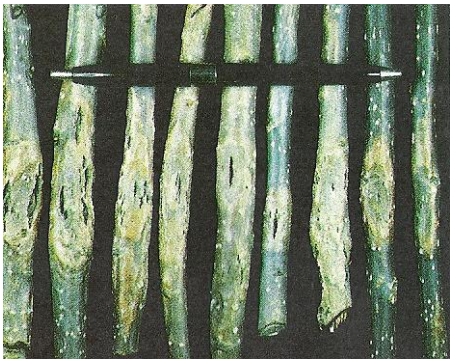
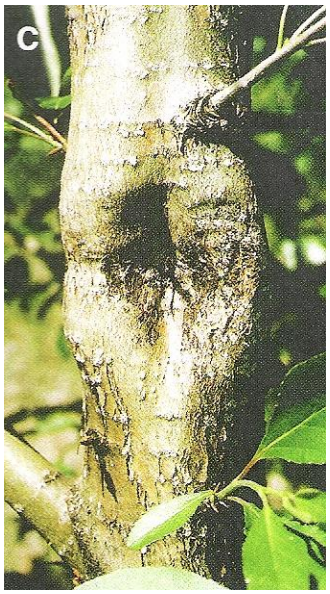


Figure 41. Several *S. musiva*-infected poplar branches showing variability in appearances (from Palmer et al., 1980).

Figure 42. Old hybrid poplar with lignification following the development of a *S. musiva* canker. The lateral thickenings illustrate the tree healing capacity of some resistant clones (from Sinclair and Lyon, 2005).



vii. Risk factors for *S. musiva*-associated cankers

The load of *S. musiva* in the poplar environment must pose as a risk factor since plantations generally increase with *S. musiva*-associated disease once it is established in that setting. Mechanisms that increase tree wound also increase the risk for cankers. For example, coppicing may allow for increased stem wounds as the new growth is proximal and subject to wind battering and consequent wound. Young trees and young shoots are at greater risk in any plantation.

Less densely spaced trees in plantations had more frequent cankers when examined after four years of initiation (Bowersox and Merrill, 1976). Increased canker formation has also been noted to correlate with increased incremental height after the

third year, suggesting that increased height was associated with decreased underfoliage or underfoliage hardiness. The latter effect was also noted by Abebe and Hart (1990). The latter authors also noted increased canker formation when borer (weevil) injury was more prevalent and also comparatively more in soils that were more enriched in phosphorus, potassium, and magnesium. Poplars in soils with relatively more iron and aluminum had fewer cankers. The latter studies did not indicate whether the association with any particular element was solely a correlation generally or whether such parameters were at the extremes of usual soil content.

Woodbury et al. (1994) examined ozone stress, and they associated increased ozone (in theory as a consequence of air pollution) with increased canker formation.

The role of water has gathered considerable attention. It is generally indicated that more disease tends to follow cool, wet weather in late spring and early summer (Newcombe et al., 1995). Bier (1961) associated increased risk for disease with drought and drying conditions. When bark turgidity was over 80%, resistance was more common. In other greenhouse experimentation, Maxwell et al. (1997) found that there was increased canker disease with water stress. LeBoldus et al. (2007), however, did not find that water stress affected poplar-*S. musiva* interactions, nor was there a correlation with root collar diameter. Among *P. balsamifera* provenances in Manitoba, *S. musiva* was more likely to cause disease in trees obtained from mesic climate zones.

What follows from all of these studies is the notion that observations in univariate analyses must be seen as just that. Findings of correlation with greater or lesser disease can only be seen as hypothesis generating findings rather than hypothesis testing findings. Multivariate analyses should be conducted in which covariables can be

assessed, since there are many variables in the field which could affect the pathogen-host interaction. Such a model for study should be encouraged. Furthermore, the above citations largely do not arise from studies of the *S. musiva-P. trichocarpa* pathosystem directly. Appropriate studies with this pathogen-host combination are needed to garner most relevance for the scenario in British Columbia.

viii. Microscopic features of *S. musiva* causing cankers

Weiland and Stanosz (2007) provided insight regarding *S. musiva* invasion in an experimental model of hybrid poplar invasion. Branches of the resistant hybrid *P. deltoides* x *P. nigra* and the susceptible hybrid *P. trichocarpa* x *P. maximowiczii* were inoculated over the leaf removal wound with *S. musiva* and thereafter observed. Within 4-6 weeks, the plant was invaded to various depths. Fungal colonization was more evident in the previously proposed susceptible hybrid which included *P. trichocarpa* parent lineage. Periderm formation as a consequence of infection was thicker in the resistant clone. As well, the necrophylactic periderm was closer to the wound for the resistant hybrid, but the susceptible hybrid developed deeper and successive responses. Fungal hyphae were present in the necrotic epidermis and outer cortex of the wound surface. Hyphae penetrated tissue external to the vascular cambium and was mainly extracellular; they were also present in vascular bundles. The observation of *S. musiva* in xylem elements shows the extent of fungal penetration and suggests potential for both longitudinal and lateral expansion of the pathogen. Although the thickness of necrophylactic periderm correlated with hybrid susceptibility, the thick periderm in itself does not confer absolute resistance.

The above is consistent with the findings of Zalasky (1978) who described the direction to occur from initial surface infection to the phloem after cell to cell migration, and thereafter for the infection to move vertically along the phloem.

The above microscopic features are similar to the disease observed when *Valsa sordida* penetrates the *P. trichocarpa* x *P. maximowiczii* hybrid and causes blackstem canker (Biggs et al., 1983). The latter suggests that up to a point, different fungi may have common mechanisms of invasion.

ix. *S. musiva* and *Populus* spp. susceptibility in North America

Although *Populus* spp. vary in susceptibility to *S. musiva* under natural and experimental conditions, Sinclair and Lyon (2005) claim that all North American poplar have some degree of susceptibility. It appears that hybrids and Euroasian species are among the most susceptible (Zalasky et al., 1968; Ostry, 1987; Mottet et al., 1991). *P. trichocarpa*, and its hybrids, also rank high in susceptibility even though natural *S. musiva* infection is not typically in the range of native *P. trichocarpa* (Mottet et al., 1991; Robison and Raffa, 1998). For example, Bier (1942) had recognized that many imported and native hybrid poplars were susceptible to canker.

Outside of the above generalizations, there are many citations of natural and experimental infection among *Populus* spp. and their hybrids. Table 7 outlines the majority of these citations but may not be totally inclusive of the experience. The use of hybrid crosses to introduce resistance genes must be balanced with the possibility that hybrids may yield less biomass (Ostry and Berguson, 1993). In general, *Ageiros* Section poplars are more resistant to *S. musiva* than are *Tacamahaca* Section poplars (Mottet et al., 1991).

Table 7. *Populus* spp. and related hybrids which have been cited in the scientific literature as being susceptible to *S. musiva* leaf spot and/or canker whether naturally or under experimental conditions. This is only a partial list; see references for further details (Thompson, 1941;Waterman, 1946; Waterman, 1954b;Bier, 1961;Filer et al., 1971;Palmer and Schipper, 1979;Long et al., 1986;Abebe and Hart, 1990;Mottet et al., 1991;Ostry and Berguson, 1993;Yang et al., 1994;Maxwell et al., 1997;Stanosz et al., 2001;Weiland et al., 2003;LeBoldus et al., 2007).

P. acuminata	*P. angulata*	*P. angustifolia*	*P. balsamifera*
P. canadensis	*P. candicans*	*P. caudina*	*P. charkoviensis*
P. deltoides	*P. fremontii*	*P. generosa*	*P. grandidentata*
P. incrassata	*P. kangitaliana*	*P. maximowiczii*	*P. monilifera*
P. nigra	*P. occidentalis*	*P. robusta*	*P. roegneriana*
P. rasumowskyana	*P. sargentii*	*P. szechuanica*	*P. simonii*
P. tamahaca	*P. tremuloides*	***P. trichocarpa***	

P. balsamifera x *P. simonii*
P. balsamifera x *P. tristis*
P. candicans x (*P. laurifolia* x *P. nigra* –commonly known as *P.* x *petrowskyana* or *P.* x *berolinensis*)
P. deltoides x *P. balsamifera* (commonly known as *P.* x *jackii*)
P. deltoides x (*P. laurifolia* x *P. nigra* –commonly known as *P.* x *petrowskyana* or *P.* x *berolinensis*)
P. deltoides x *P. maximowiczii*
P. deltoides x *P. nigra* (commonly known as *P.* x *euroamericana*)
P. deltoides x ***P. trichocarpa***
P. laurifolia x *P. nigra* (commonly known as *P.* x *petrowskyana* or *P.* x *berolinensis*)
P. maximowiczii x (*P. deltoides* x ***P. trichocarpa***)
P. maximowiczii x (*P. laurifolia* x *P. nigra* –commonly known as *P.* x *petrowskyana* or *P.* x *berolinensis*)
P. maximowiczii x ***P. trichocarpa***
P. nigra x (*P. laurifolia* x *P. nigra* –commonly known as *P.* x *petrowskyana* or *P.* x *berolinensis*)
P. nigra var. charkowiensis x *P. nigra* Incrassata
P. nigra var. charkowiensis x *P. trichocarpa*
P. nigra x ***P. trichocarpa***
P. nigra x *P. maximowiczii*
P. simonii x (*P. laurifolia* x *P. nigra* –commonly known as *P.* x *petrowskyana* or *P.* x *berolinensis*)

x. Silviculture practice and *S. musiva*

Silviculture practices are anticipated to affect the presence and spread of *S. musiva* in British Columbia. The following are examples of variables that could prove of relevance:

a) coppicing – this practice is associated with increased young stem canker in regions where the technique is widely adopted. This technique, however, is not ascribed to largely in this province.

b) limiting stress factors in the plantation – factors such as water exposure, relative humidity, rainfall, fertilizer, spacing, and ground clearing around trees may be relevant.

c) international importation of susceptible poplar and hybrids from outside of the continent.

d) importation from greenhouses or plantations where *S. musiva* is endemic in other geographic areas of North America, e.g., central Canada and northeast U.S.

e) moving plants and cuttings from endemic plantations in the province to another plantation in the province.

f) plantation farming – close proximity of susceptible poplar in large groups will facilitate plant to plant spread.

g) repetitive use of silviculture site and poplars – endemic propagation of fungus when the same growth site is being used repetitively. Rotations with the same susceptible poplars and their hybrids can maintain the source for reinfection.

It is important to acknowledge in anticipation of the above that attempts to sterilize poplar cuttings carrying *S. musiva* have not often been successful (Boyce, 1954). Therefore, mobilization of trees and cuttings from infected to non-infected areas should be regarded with caution for disease spread. Indeed, perhaps such a phenomenon has already occurred in relocating *S. musiva* to the Lower Fraser Valley (Beauseigle et al., 2010). One would not appreciate having the same problem with canker in British Columbia plantations as has been described at times in Ontario, Quebec, and northcentral United States (Strobl and Fraser, 1989).

xi. Treatment and prevention

Active treatment of *S. musiva* infection, whether for leaf spot or canker, is also an area for research since not much effort has been published in this regard. Cankers in particular may be theoretically amenable to some form of local treatment early in the canker formation stage, especially if combined with a previously existing and relatively resistant poplar clone that has the ability to heal diseased bark.

Silviculture practice modalities, as for *S. populicola*, merit consideration. These include removal leaf debris in the late fall, use of resistance poplar clones, and assurance of disease-free stock. The removal of leaf debris by cultivation in the spring did not reduce *S. musiva* canker in one hybrid poplar study (Ostry,1987). The early harvest and replacement of diseased clones could also be of assistance. Silviculturists should choose locations which are optimal for growth and yet less likely to bear the hazard for tree stress. Quarantine for tree imports must also be considered.

The enhancement of tree resistance to *S. musiva* is currently attracting considerable attention. Whether through natural selection, hybrid formation, or genetic or other biotechnical manipulation, a more fit poplar may be found. For example, one could look at selecting optimally resistant *P. trichocarpa* provenances among the numerous provenances that may exist within the province. In addition, one could also look at crossing provenances to further select for optimal resistance genes. Of interest, such an approach was already studied by Michiels et al. (2000) in Europe for assessing North American provenances in regard to rust susceptibility, form, and vigor. Studies in eastern Canada have looked at breeding for resistance among other poplars (Mottet and Perinet, 2000). Newcombe and Ostry (2001) provide evidence for a recessive gene pattern of

inheritance for *S. musiva* resistance in *P. trichocarpa* x *P. deltoides* hybrids. In the latter experiments, *P. trichocarpa* and the F1 hybrids were susceptible to canker, but *P. deltoides* and 10 of 70 F2 hybrids were resistant. Some have attempted to derive resistant hybrids through growth from tissue culture, and they achieved some success; an in vitro predictor for such resistance could not be determined however – in particular, the leaf disk bioassay was not predictive for selecting resistant clones (Ostry and Skilling, 1988; Ostry and Ward, 2003).

Biological approaches to *S. musiva* control have been published. In one example, a *Streptomyces* bacterium has been applied to trees though aerosol in order to use the power of disease-suppressing strains to combat the fungus (Gyenis et al. 2003). In another similar approach, the fungus *Phaeotheca dimorphospora* is applied in order to inhibit *S. musiva* by means of antifungal agent secretion. Both of these are of interest and some benefit, but they have gathered more general interest rather than practical use (Yang et al., 1993; Yang et al., 1994).

Gyenis et al. (2003) have also shown that detergent application to leaves can be a deterrent for leaf spot. Chemical applications can also reduce *Septoria* infection (Waterman, 1954b; Ostry, 1987). Zalasky (1978) suggests that application of a fungicide should occur when the leaves first begin to show leaf spots, especially in dry climate.

Genetic engineering of poplars is enticing to scientists as a method to enhance resistance. In this regard, for example, the transformation of poplar clones with antimicrobial peptide-producing genes has already been attempted with some success (Liang et al., 2002). Such studies are in their infancy. The role of endogenous poplar antifungal compounds deserves study (Shain and Miller, 1982).

Importation of infected clones is a concern. Methods to decontaminate potentially infected cuttings will be of value (Waterman and Aldrich, 1952; Waterman, 1954a; Waterman, 1954b). Likewise, it is possible that exportation of North American, and British Columbian, biologicals could lead to infection in other parts of the world (Riker, 1961). Surface sterilization of poplar cuttings has some benefit in reducing the fungal load, e.g., mercuric chloride with and without alcohol, but it is nearly impossible to totally remove fungal contamination without seriously injuring the cutting. The resinous scales of the buds allow the fungal spores to adhere tightly.

V. Summary

P. trichocarpa is emerging in its importance both for the potential to this province's economy and for its value to the understanding of plants, trees, and *Populus* spp. generally. The main break through has been the sequencing of its genome which will now allow for an immediate and dramatic growth in black cottonwood research. The spin-offs are imminently considerable. Such progress will contribute to the insight required to better assess the disease caused by *Septoria* spp. There is much to be considered in the applications of these future studies to the value of black cottonwood biomass.

In contrast to the above, the science relating to *Septoria* spp., while emerging, is in its infancy. Whether for *S. populicola* or *S. musiva*, relatively little is known about mechanisms of pathogenesis, whether at the molecular or gross pathology levels. There are many other unknowns in the understanding of *Septoria* spp. diseases. Of concern is

the recent emergence of *S. musiva* among poplars in British Columbia. The insight into the latter is at this time embryonic.

The *Septoria* spp.-*P. trichocarpa* pathosystem requires much further investigation. As the science for either fungus or tree progresses, we must thereafter look intimately at the way in which these species interact and indeed allow for disease. Given the potential growth in the underderstanding of black cottonwood at the genetic level, we should be able to exploit the latter to understand better how host and pathogen interact at the microscopic and molecular strata. We now have the potential to examine these interactions as model systems which might be cross-applicable to other tree pathology.

British Columbia is well positioned to provide leadership in these areas as there are already a number of investigators who are looking at various aspects of the above. Given the prominence of *P. trichocarpa* in this province, and given the advances from local scientists at the University of British Columbia among others from the National Forestry Centre and industry, there is great opportunity to advance the science.

VI. Recommendations for Future Practice and Research

As illustrated in the news brief from Natural Resources Canada (Keiran, 2008), local scientists are already activated to further consider *S. musiva* in the Fraser Valley of British Columbia, and some prevention strategies are under way. Feau et al. (2007b) have looked at the *Melampsora – P. trichocarpa* pathosystem and have provided some guidance on future endeavours in what they call the "model pathogens for a model tree". It is apparent from the latter that the *Septoria – P. trichocarpa* pathosystem will have

many parallels and that either future practice or future research will benefit from the same insights as detailed by Feau et al. (2007b).

The following points serve as a brief highlight for future endeavours which might be considered given the knowledge base that is detailed in this review:

◦ enhance the general understanding of *P. trichocarpa* responses to pathogens, especially fungal, and especially in regards to the immune system if any

◦ better understanding of hybrid genetics and the determinants of resistance

◦ further the understanding of the genetic variability of *P. trichocarpa*

◦ better defining the role of *P. trichocarpa* as a carbon sink

◦ further studies for determining the diversity of *S. musiva*

◦ determining the virulence factors of *S. musiva*

◦ fingerprinting local isolates of *S. musiva* and examining their relationship to isolates from other North American regions or elsewhere

◦ conducting broader surveys for *S. musiva* among native trees in British Columbia, but especially among *P. trichocarpa*

◦ surveying year-to-year frequency of infection of *S. musiva* among plantation trees and determing factors that may lead to variation

◦ determining whether there are significant differences for infection among *P. trichocarpa* which are obtained from a common geographic zone but from varying topographies within that zone, e.g. from trees in a xeric versus mesic environment from that same geography

◦ conduct a multivariate analysis of risk factors for *S. musiva* infection among *P. trichocarpa* and/or its hybrids

◦ examine the differential susceptibility of *P. trichocarpa* provenances in British Columbia to *S. musiva* disease

◦ further work in specifically understanding the *S. musiva – P. trichocarpa* pathosystem in regards to interactive genetics, immunity, spread, treatment, and prevention

◦ better understanding of the microscopic features of *S. musiva* in its cause of leaf spot and canker

◦ developing a model system, either in the greenhouse or laboratory, for more accurately predicting which trees are more or less susceptible to *S. musiva*

◦ tracing the history of trees that were grown in the same region/plantations of British Columbia where *S. musiva* has been found

◦ survey for the presence of *S. musiva* disease among other plant hosts in the same geography as *P. trichocarpa*

◦ enhancing strategies for canker prevention

◦ defining techniques for early canker treatments

Where *S. musiva* is named in any one of the above areas for future concern, the fungus could easily be replaced in name by *S. populicola*

VII. Acknowledgements

I thank Dr. Richard Hamelin and Stephanie Beauseigle for their academic input in providing the stimulus for this review. I acknowledge Dr. Brenda Callan for her clarification of the findings in her paper (Callen et al., 2007) and in the Forest Pathology Herbarium index.

VIII. References

Note: References in which studies have been conducted mainly in British Columbia, or in which tree clones from British Columbia are used, have been highlighted by **bolding**. Some details of important references are highlighted by discourse coloured in green.

Abebe G, Hart JH. (1990) The relationship of site factors to the incidence of *Cytospora* and *Septoria* cankers and poplar and willow borer in hybrid poplar plantations. USDA Forest Service Gen Tech Rep, NC-140.

Abrahamson LP, White EH, Nowak CA. (1990) Evaluating hybrid poplar clonal growth potential in a three-year-old genetic selection field trial. Biomass 21:101-114.

Alfieri Jr. SA. (1991) *Septoria* leaf spot of blueberry. Plant Pathology Circular No. 340, Florida Department of Agriculture and Consumer Services, Contribution #662.

Balatinecz J, Mertens P, De Boever L, Yukun H, Jin J, Van Acker J. (2010) Properties, processing, and utilization. Chapter 10. *In*: Poplars and Willows in the World. International Poplar Commission Working Paper IPC/9-10, Rome, Italy. Forestry Department, Food and Agriculture Organization of the United Nations.

Bassman JH, Zwier JC. (1991) Gas exchange characteristics of *Populus trichocarpa*, *Populus deltoides* and *Populus trichocarpa* X *P. deltoides* clones. Tree Physiol 8:145-159.

Beauseigle S, Feau N, Hamelin RC. (2010) Poplar leaf spot and canker caused by *Septoria musiva* in British Columbia: risk assessment. Report to Pest and Forest Management, Province of British Columbia, April, 2010. Contemporary report identifying the presence of *S. musiva* in a Fraser Valley plantation and among natural populations of *P. trichocarpa* in the same region.

Bier JE. (1939) *Septoria* canker of introduced and native hybrid poplars. Can J Res [C] 17:195-204. Sentinel research on *Septoria* in Canada. While this study was from Bier in his position in Ottawa, eventually Bier became integral to studies in British Columbia.

Bier JE. (1942) Forest pathology in British Columbia. Pulp Paper Magazine Canada 43:528-530. A view on the state of the art some 60 years ago.

Bier JE. (1961) The relation of bark canker to the development of canker diseases caused by native, facultative parasites. VI. Pathogenicity studies of *Hypoxylon pruinatum* (Klotzsch) Cke. and *Septoria musiva* Pk. on species of *Acer, Populus,* and *Salix*. Can J Bot 39:1555-1561.

Biggs AR, Davis DD, Merrill W. (1982) Histopathology of cankers on *Populus* caused by *Cytospora chrysosperma*. Can J Bot 61:563-574.

Biggs AR, Merrill W, Davis DD. (1984) Response of bark tissues to injury and infection. Can J For Res 14:351-356.

Bjorkman E. (1964) 7. Breeding for resistance to disease in forest trees. FAO Corporate Document Repository, FAO/IUFRO meeting on forest genetics. Accessed in: http://www.fao.org/docrep/03650e/03650e08.htm (November, 2010) Unasylva 18(2-3).

Boes TK, Strauss SH. (1994) Floral phenology and morphology of black cottonwood, *Populus trichocarpa* (Salicaceae). Amer J Bot 81:562-567. A key paper on phenology.

Bowersox TW, Merrill W. (1976) Stand density and height increment affect incidence of *Septoria* canker in hybrid poplar. Plant Dis Rep 60:835-837.

Boyce JS. (1954) Introduction of exotic trees: dangers from disease pests. FAO Corporate Document Repository. Accessed in: http://www.fao.org/docrep/x5370e/x5370e02.htm (November, 2010) Unasylva 8(1).

Brown-Skrobot S, Brown LR. (1984) Ethylene and carbon monoxide production in *Septoria musiva*. Developments Industrial Microbiology 25:749-755.

Callan BE. (1998) Diseases of *Populus* in British Columbia: a diagnostic manual. Cdn For Serv, Natural Resources Canada. A beautiful diagnostic manual for poplar disease in British Columbia.

Callan BE, Leal I, Foord B, Dennis JJ, van Oosten C. (2007) *Septoria musiva* isolated from cankered stems in hybrid poplar stool beds, Fraser Valley, British Columbia. Pac NW Fungi 2:1-9. A very important and contemporary paper on the presence of *S. musiva* in British Columbia.

Carbon LW. (1972) Fungicidal control of poplar leaf spots in Alberta and Saskatchewan. Can Plant Dis Surv 52:99-101.

Cellerino GP. (1999) Review of fungal diseases in poplar. FAO Corporate Document repository, Food and Agriculture Organization of the United Nations, Rome. Accessed in: http://www.fao.org/docrep/004/ac492e/ac492e00.htm (November, 2010)

Ceulemans R, Scarascia-Mugnozza G, Wiard BM, Braatne JH, Hinckley TM, Stettler RF, Isebrands JG, Heilman PE. (1992) Production physiology and morphology of *Populus* species and their hybrids grown under short rotation. I. Clonal comparisons of 4-year growth and phenology. Can J For Res 22:1937-1948. Included some clones from British Columbia.

Cisneros HA, Belanger L, Gee WY, Watson PA, Hatton JV. (2000) Wood and fiber properties of hybrid poplars from southern British Columbia. Tappi 83:60.

Cooke JEK, Rood SB. (2007) Trees of the people: the growing science of poplars in Canada and worldwide. Can J Bot 85:1103-1110.

Cooper DT, Filer Jr. TH. (1976) Resistance to *Septoria* leaf spot in eastern cottonwood. Plant Dis. Rep. 60:812-814.

DaQiang W, Cheng C, Guo W, Yan X. (2009) Genome-wide analysis of *NBS*-encoding disease resistance genes in *Populus trichocarpa*. Scientia Silvae Sinicae 45:152-157.

DeBell DS. (1975) Short-rotation culture of hardwoods in the Pacific Northwest. Iowa State J Res 49:345-352.

DeBell DS, Radwan MA. (1979) Growth and nitrogen relations of coppiced black cottonwood and red alder in pure and mixed plantings. Bot Gaz 140(Suppl.):97-101.

DeBell DS. (1990) Black Cottonwood Populus trichocarpa Torr. & Gray. Accessed from http://www.na.fs.fed.us/spfo/pubs/silvics_manual/volume_2/populus/trichocarpa.htm (accessed November, 2010) A useful concise review.

Demerritt Jr. ME. (1990) Poplar hybrids. *In*: Silvics Manual, Volume 2. Retrieved from http://www.na.fs.fed.us/pubs/silvics_manual/volume_2/populus/populus/htm (accessed November, 2010)

deVries SMG. (2001) Conservation of natural ecosystems of poplar and willow. Forest Chronicle 77:255-257.

Dickmann DI, Kuzovkina J. (2008) Poplars and willows of the world, with emphasis on silviculturally important species. Chapter 2. *In*: Poplars and Willows in the World. International Poplar Commission Working Paper IPC/9-2, Rome, Italy. Forestry Department, Food and Agriculture Organization of the United Nations.

Dillen SY, Rood SB, Ceulemans R. (2010) Growth and physiology. *In*: Genetics and Genomics of *Populus*. Springer Publishing, New York, NY.

Douglas CJ, DiFazio SP. (2010) The *Populus* genome and comparative genomics. *In*: Genetics and Genomics of *Populus*. Springer Publishing, New York, NY.

Dunlap JM, Braatne JH, Hinckley TM, Stettler RF. (1993) Intraspecific variation in photosynthetic traits of *Populus trichocarpa*. Can J Bot 71:1304-1311.

Dunlap JM, Heilman PE, Stettler RF. (1995) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. VIII. Leaf and crown morphology of native *P. trichocarpa* clones from four river valleys in Washington. Can J For Res 25:1710-1724.

Dunlap JM, Stettler RF. (1996) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. IX. Phenology and *Melampsora* rust incidence of native

black cottonwood clones from four river valleys in Washington. For Ecol Manage 87:233-256.

Dunlap JM, Stettler RF. (1998) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. X. Trait correlations in young black cottonwood from four river valleys in Washington. Trees 13:28-39.

Farrar JL. (1995) Black cottonwood. *In*: Trees in Canada. Canadian Forest Service and Fitzhenry & Whiteside Ltd., Markham, Canada. pp.338-339.

Feau N, Hamelin RC, Vandecasteele C, Stanosz GR, Bernier L. (2005a) Genetic structure of *Mycosphaerella populorum* (anamorph *Septoria musiva*) populations in north-central and northeastern North America. Phytopathol 95:608-616.

Feau N, Weiland JE, Stanosz GR, Bernier L. (2005b) Specific and sensitive PCR-based detection of *Septoria musiva*, *S. populicola* and *S. populi*, the causes of leaf spot and stem canker on poplars. Mycol Res 109:1015-1028.

Feau N, Hamelin RC, Bernier L. (2006a) Attributes and congruence of three molecular data sets: inferring phylogenies among *Septoria*-related species from woody perennial plants. Mole Phylogenet Evol 40:808-829.

Feau N, Jacobi V, Hamelin RC, Bernier L. (2006b) Screening of ESTs from *Septoria musiva* (teleomorph *Mycosphaerella populorum*) for detection of SSR and PCR-RFLP markers. Molecular Ecol Notes 6:356-358.

Feau N, Hamelin RC, Bernier R. (2007a) Variability of nuclear SSU-rDNA group introns within *Septoria* species: incongruence with host sequence phylogenies. J Mol Evol 64:489-499.

Feau N, Joly DL, Hamelin RC. (2007b) Poplar leaf rusts: model pathogens for a model tree. Can J Bot 85:1127-1135.

Feau N, Mottet MJ, Perinet P, Hamelin RC, Bernier L. (2010). Recent advances related to poplar leaf spot and canker caused by *Septoria musiva*. Can J Plant Pathol 32:122-134.
The most contemporary and extensive review of this topic.

Filer TH, McCracken FI, Mohn CA, Randall WK. (1971) *Septoria* canker on nursery stock of *Populus deltoides*. Plant Dis Rep 55:460-463.

Forest Pathology Herbarium, Pacific Forestry Centre, Victoria, B.C.
http://www.pfc.cfs.nrcan.gc.ca/biodiversity/herbarium/index_e.html
(accessed November, 2010) A underrecognized resource for plant scientists in British Columbia.

Gilchrist EJ, Haughn GW, Ying CC, Otto SP, Zhuang J, Cheung D, Hamberger B, Aboutorabi F, Kalynyak T, Johnson L, Bohlmann J, Ellis BE, Douglas CJ, Cronk QCB. (2006) Use of Ecotilling as an efficient SNP discovery tool to survey genetic variation in wild populations of *Populus trichocarpa*. Molecular Ecology 15:1367-1378.

Gordon JC. (2001) Poplars: trees of the people, trees of the future. For Chronicle 77:217-219.

Gornall JL, Guy RD. (2007) Geographic variation in ecophysiological traits of black cottonwood (*Populus trichocarpa*). Can J Bot 85:1202-1213. A paper which strikes to the heart of understanding the provenances within British Columbia.

Groover AT, Nieminen K, Helariutta Y, Mansfield SD. (2010) Wood formation in *Populus*. In: Jansson S, et al. Genetics and Genomics of *Populus*. Springer, New York, NY. 1st Edn.

Gyenis L, Anderson NA, Ostry ME. (2003) Biological control of *Septoria* leaf spot disease of hybrid poplars in the field. Plant Dis 87:809-813.

Hamel LP, Miles GP, Samuel MA, Ellis BE, Seguin A, Beaudoin N. (2005) Activation of stress-responsive mitogen-activated protein kinase pathways in hybrid poplar (*Populus trichocarpa* X *Populus deltoides*). Tree Physiology 25:277-288.

Harrington CA, DeBell DS. (1984) Effects of irrigation, pulp mill sludge, and repeated coppicing on growth and yield of black cottonwood and red alder. Can J For Res 14:844-849.

Heilman PE, Peabody Jr DV, DeBell DS, Strand RF. (1972) A test of close-spaced, short-rotation culture of black cottonwood. Can J For Res 2:456-459.

Heilman PE, Ekuan G. (1979) Effect of planting stock length and spacing on growth of black cottonwood. Forest Sci 35:439-443.

Heilman P, Peabody Jr. DV. (1981) Effect of harvest cycle and spacing on productivity of black cottonwood in intensive culture. Can J For Res 11:118-123.

Heilman PE, Stettler RF. (1985a) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. II. Biomass production in a 4-year plantation. Can J For Res 15:384-388.

Heilman PE, Stettler RF. (1985b) Mixed, short-rotation culture of red alder and black cottonwood: growth, coppicing, nitrogen fixation, and allelopathy. Forest Sci 31:607-616.

Heilman PE, Stettler RF. (1986) Nutritional concerns in selection of black cottonwood and hybrid clones for short rotation. Can J For Res 16:860-863.

Heimburger C. (1936) Report on poplar hybridization. Forest Chronicle 12:285-290. An early Canadian report.

Hildebrand PD, Renderos WE, Filmore SAE. (2010) Severity of *Septoria* leaf spot and stem canker and leaf rust in lowbush blueberry fields pruned by mowing or burning. Canadian Plant Disease Survey, The Canadian Phytopathological Society. pp. 155-157.

Howe GT, Hackett WP, Furnier GR, Klevorn RE. (1995) Photoperiodic responses of a northern and southern ecotype of black cottonwood. Physiol Plant 93:695-708.

Johnson JD, Kim Y. (2005) The role of leaf chemistry in *Melampsora medusae* infection in hybrid poplar: effects of leaf development and fungicide treatment. Can J For Res 35:763-771.

Jones RK, Benson DM. (2001) Diseases of Woody Ornamentals and Trees in Nurseries. APS Press, The American Phytopathological Society, St. Paul, Minnesota.

Kang KY, Bradic S, Avramidis S, Mansfield SD. (2007) Kiln-drying lumber quality of hybrid poplar clones. Holzforschung 61:65-73.

Keiran M. (2008) Pathogen`s presence prompts prevalence probe. Information forestry, Natural Resources Canada. Accessed in: http://cfs.nrcan.gc.ca/news/656 (December, 2010).

Klepzig KD, Robison DJ, Smalley EB, Raffa KF. (1997) Effects of feeding by two folivorous arthropods on susceptibility of hybrid poplar clones to a foliar pathogen. The Great Lakes Entomologist 30:99-104.

Klinka K, Worrall J, Skoda L, Varga P. (2000) *Populus trichocarpa*. *In*: The Distribution and Synopsis of Ecological and Silvical Characteristics of Tree Species of British Columbia's Forests. Canadian Cartographics Ltd., Coquitlam, B.C. A compendium of British Columbia's forest resources.

Kohler A, Rinaldi C, Duplessis S, Baucher M, Geelen D, Duchaussoy F, Meyers BC, Boerjan W, Martin F. (2008) Genome-wide identification of *NBS* resistance genes in *Populus trichocarpa*. Plant Mol Biol 66:619-636.

Krupinsky JM. (1989) Variability in *Septoria musiva* in aggressiveness. Phytopathol 89:413-416.

LeBoldus JM, Blenis PV, Thomas BR. (2007) Evaluating the interaction between genotype and water stress in the hybrid poplar – *Septoria musiva* pathosystem. Can J Bot 85:1098-1102.

LeBoldus JM, Blenis PV, Thomas BR. (2008) Clone by isolate interaction in the hybrid poplar-*Septoria musiva* pathosystem. Can J For Res 38:1888-1896.

LeBoldus JM, Blenis PV, Thomas BR, Feau N, Bernier L. (2009) Susceptibility of *Populus balsamifera* to *Septoria musiva*: a field study and greenhouse experiment. Plant Dis 93:1146-1150.

Liang H, Catranis CM, Maynard CA, Powell WA. (2002) Enhanced resistance to the poplar fungal pathogen, *Septoria musiva*, in hybrid poplar clones transformed with genes encoding antimicrobial peptides. Biotechnol. Lett. 24:383-389.

Lo MH, Abrahamson LP, White EH, Manion PD. (1995) Early measure of basal area and canker disease predict growth potential of some hybrid poplar clones. Can J For Res 25:1113-1118.

Long R, Bowersox TW, Merrill W. (1986) Artificial inoculation of *Populus* hybrids with *Septoria musiva*. Can J For Res 16:405-407.

Luley CJ, McNabb HS. (1989) Ascospore production, release, germination, and infection of *Populus* by *Mycosphaerella populorum*. Phytopathology 79:1013-1018. Important details of the life cycle.

Luley CJ, McNabb HS. (1991) Estimation of seasonal ascospore production of *Mycosphaerella populorum*. Can J For Res 21:1349-1353.

Major IT, Constabel CP. (2007) Shoot-root defense signaling and activation of root defense by leaf damage in poplar. Can J Bot 85:1171-1181.

Marron N, Gielen B, Brignolas F, Jian G, Johnson JD, Karnosky DF, Polle A, Scarascia-Mugnozza G, Schroeder WR, Ceulemans R. (2008) Abiotic stress. Chapter 7. *In*: Poplars and Willows in the World. International Poplar Commission Working Paper IPC/9-7, Rome, Italy. Forestry Department, Food and Agriculture Organization of the United Nations.

Maxwell DL, Kruger EL, Stanosz GR . (1997) Effects of water stress on colonization of poplar stems and excised leaf disks by *Septoria musiva*. Phytopathology 87:381-388.

McCamant T, Black RA. (2000) Cold hardiness in coastal, montane, and inland populations of *Populus trichocarpa*. Can J For Res 30:91-99.

Michiels B, Steenackers M, Steenackers V, Van Slycken J. (2000) A long-term planned *P. trichocarpa* program, included domestication. Abstracts of the 21st Session of the International Poplar Commission. FAO Corporate Document Repository. Accessed in: http://www.fao.org/docrep/MEETING/004/AC351E/AC351E04.htm (December, 2010)

Miranda M, Ralph Sg, Meilway R, White R, Heath MC, Bohlmann J, Constabel CP. (2007) The transcriptional response of hybrid poplar (*Populus tricocarpa* X *P. deltoides*) to infection by *Meampsora medusae* leaf rust involves induction of flavonoid pathway genes leading to the accumulation of proanthocyanidins. Molecular Plant-Microbe Interactions 20:816-831.

Mohamed R, Meilan R, Ostry ME, Michler CH, Strauss SH. (2001) Bacterio-opsin gene overexpression fails to elevate fungal disease resistance in transgenic poplar (*Populus*). Can J For Res 31:268-275.

Mohn, CA. (1969) A study of the genetic control of shoot growth patterns in *Populus trichocarpa*. Submitted as a thesis to the Graduate School of the University of Minnesota, USA.

Mottet MJ, Bussières G, Vallée G. (1991) Test précoce pour l'évaluation de la sensibilité de peupliers hybrides au chancre septorien. Forest Chronicle 67 :411-416.

Mottet MJ, Perinet P. (2000) Breeding for resistance to *Septoria* canker in Quebec, Canada. Abstracts of the 21st Session of the International Poplar Commission. FAO Corporate Document Repository. Accessed in: http://www.fao.org/docrep/MEETING/004/AC351E/AC351E04.htm (December, 2010)

Mullick DB. (1977) The non-specific nature of defense in bark and wood during wounding, insect and pathogen attack. Recent Adv Phytochem 11:395-441.

Newcombe G, Chastagner GA, Callan BE, Ostry ME. (1995) An epidemic of *Septoria* leaf spot on *Populus trichocarpa* in the Pacific Northwest in 1993. Plant Dis 79:212. An abstract which includes the finding of extensive leaf spot in British Columbia.

Newcombe G, Bradshaw HD. (1996) Quantitative trait loci conferring resistance in hybrid poplar to *Septoria populicola*, the cause of leaf spot. Can J For Res 26:1943-1950.

Newcombe G. (1998) A review of exapted resistance to diseases of *Populus*. Eur J For Path 28:209-216.

Newcombe G, Ostry M. (2001) Recessive resistance to *Septoria* stem canker of hybrid poplar. Phytopathol 91:1081-1084.

Newcombe G. (2005) Genes for parasite-specific, nonhost resistance in *Populus*. Phytopathology 95:779-783. Includes samples from British Columbia.

Nimchuk Z, Eulgem T, Holt III BF, Dangl JL. (2003) Recognition and response in the plant immune system. Annu Rev Genet 37:579-609.

Ostry ME, McNabb HS. (1985) Susceptibility of *Populus* species and hybrids to disease in the North Central United States. Plant Dis 69:755-757.

Ostry M. (1987) Biology of *Septoria musiva* and *Marssonina brunnea* in hybrid *Populus* plantations and control of *Septoria* canker in nurseries. Eur J For Path 17:158-165.

Ostry ME, Skilling DD. (1988) Somatic variation in resistance of *Populus* to *Septoria musiva*. Plant Dis 72:724-727.

Ostry ME, Wilson LF, McNabb HS, Moore LM. (1989) A Guide to Insect, Disease, and Animal Pests of Poplars. United States Dept Agric, For Serv, Agriculture Handbook 677.

Ostry ME, Berguson WE. (1993) Selecting hybrid poplars to reduce disease risk may also reduce biomass yield. Tree Planters' Notes 44:128-131.

Ostry ME, Ward KT. Field performance of *Populus* expressing somaclonal variation in resistance to *Septoria musiva*. Plant Sci 164:1-8.

Palmer MA, Schipper AL. (1979) Resistance of poplars to *Septoria* leaf spot in relation to *Septoria* canker resistance. Phytopathology 69:1041.

Palmer MA, Schipper Jr AL, Ostry ME. (1980) How to identify and control *Septoria* leaf spot and canker of poplar. Technical reference, North Central For Exp Stn, St. Paul, MN. A short technical reference but with colour photos of plant pathology.

Pojar J, MacKinnon A. (1994) Black cottonwood. *In*: Plants of Coastal British Columbia. BC Ministry of Forests and Lone Pine Publishing, Vancouver, BC.

Ralph SG, Chun HJE, Cooper D, Kirkpatrick R, Kolosova N, Gunter L, Tuskan GA, Douglas CJ, Holt RA, Jones SJM, Marra MA, Bohlmann J. (2008) Analysis of 4,664 high-quality sequence-finished poplar full-length cDNA clones and their utility for the discovery of genes responding to insect feeding. BMC Genomics 9:57.

Rathjen JP, Moffett P. (2003) Early signal transduction events in specific plant disease resistance. Curr Opin Plant Biol 6:300-306.

Richardson J, Cooke JEK, Isebrands JG, Thomas BR, Van Rees KCJ. (2007) Poplar research in Canada – a historical perspective with a view to the future. Can J Bot 85:1136-1146. A state of the art view on activity in Canada.

Ridge CR, Hinckley TM, Stettler RF, Van Volkenburgh E. (1986) Leaf growth characteristics of fast-growing poplar hybrids *Populus trichocarpa* x *P. deltoides*. Tree Physiol 1:209-216.

Riemenschneider DE, McMahon BG, Ostry ME. (1994) Population-dependent selection strategies needed for 2-year-old black cottonwood clones. Can J For Res 24:1704-1710. Used materials from British Columbia.

Riemenschneider DE, Berguson WE, Dickmann DI, Hall RB, Isebrands JG, Mohn CA, Stanosz GR, Tuskan GA. (2001) Poplar breeding and testing strategies in the north-central U.S.: demonstration of potential yields and consideration of future research needs. Forest Chronicle 77:245-253.

Riker AJ. (1961) Internationally dangerous tree diseases. FAO Corporate Document Repository. Unasylva 15(2). Accessed in: http://www/fao.prg/docrep/x5399e/x5399e06.htm (November, 2010)

Robison DJ, Raffa KF. (1998) Productivity, drought tolerance and pest status of hybrid *Populus*: tree improvement and silvicultural implications. Biomass Bioenergy 14:1-20.

Rood SB, Kalischuk AR, Polzin ML, Braatne JH. (2003a) Branch propagation, not cladoptosis, permits dispersive, clonal reproduction of riparian cottonwoods. For Ecol Manage 186:227-242.

Rood SB, Braatne JH, Hughes FMR. (2003b) Ecophysiology of riparian cottonwoods: stream flow dependency, water relations and restoration. Tree Physiol 23:1113-1124.

Royle DJ, Ostry ME. (1995) Disease and pest control in the bioenergy crops poplar and willow. Biomass Bioenergy 9:69-79.

Sedjo RA. (2001) The role of forest plantations in the world's future timber supply. Forest Chronicle 77:221-225.

Semple KE, Vaillant MH, Kang KY, Oh SW, Smith GD, Mansfield SD. (2007) Evaluating the suitability of hybrid poplar clones for the manufacture of oriented strand boards. Holzforschung 61:430-438.

Shain L, Miller JB. (1982) Pinocembrin: an antifungal compound secreted by leaf glands of eastern cottonwood. Phytopathol 72:877-880.

Silen RR. (1947) Comparative growth of hybrid poplars and native northern black cottonwoods. Forest Research Notes No. 35, Pacific Northwest Forest Experiment Station, USA.

Sillick JM, McNabb Jr. HS, Thornburg RW. (1989) Extracellular proteases of *Septoria musiva*. Phytopathol 79:1005.

Sinclair WA, Lyon HH. (2005) Diseases of Trees and Shrubs, Second Edition. Cornell University Press, Ithaca, NY.

Slavov GT, Zhelev P. (2010) Salient biological features, systematics, and genetic variation of *Populus*. *In*: Genetics and Genomics of Populus. Springer Publishing, New York, NY.

Smit BA. (1988) Selection of flood-resistant and susceptible seedlings of *Populus trichocarpa* Torr. & Gray. Can J For Res 18:271-275.

Smith JHG, Haddock PG, Hancock WV. (1956) Topophysis and other influences on growth of cuttings from black cottonwood and Carolina poplar. J Forest 54:471-472. Further insights in regards to black cottonwood in British Columbia.

Smith JHG. (1957) Some factors indicative of site quality for black cottonwood (*Populus trichocarpa* Torr. & Gray) J Forest 55:578-580. A key paper for those interested in the history of black cottonwood in British Columbia.

Smith JHG, Blom G. (1966) Decade of intensive cultivation of poplars in British Columbia shows need for long-term research to reduce risks. Forest Chronicle 42:359-376. A then timely review of the concern regarding poplar cultivation in British Columbia.

Spielman LJ, Hubbes M. (1984) Characterization of *Septoria musiva* isolates from Ontario and the United States. Phytopathol 74:840.

Spielman LJ, Hubbes M, Lin D. (1986) *Septoria musiva* on hybrid poplar in southern Ontario. Plant Dis 70:968-971.

Stanosz GR, Stanosz JC, Rousseau RJ. (2001) Hybrid poplar stem cankers caused by *Mycosphaerella populorum* in Kentucky, USA. New Dis Rep 4:3.

Stanton BJ. (2009) The domestication and conservation of *Populus* genetic resources. Chapter 4a. *In*: Poplars and Willows in the World. International Poplar Commission.

Stanton BJ, Neale DB, Li S. (2010) *Populus* breeding: from the classical to the genomic approach. *In*: Genetics and Genomics of *Populus*. Springer Publishing, New York, NY.

Stettler RF, Fenn RC, Heilman PE, Stanton BJ. (1988) *Populus trichocarpa* x *Populus deltoides* hybrids for short rotation culture: variation patterns and 4-year field performance. Can J For Res 18:745-753.

Strobl S, Fraser K. (1989) Incidence of *Septoria* canker of hybrid poplars in eastern Ontario. Can Plant Dis Surv 69:109-111.

Tauer CG. (1975) Competition between selected black cottonwood genotypes. Silvae Genetica 24:2-3.

Thomas GP, Podmore DG. (1953) Studies in forest pathology. XI. Decay in black cottonwood in the middle Fraser region, British Columbia. Can J Bot 31:675-692.

Thompson GE. (1941) Leaf-spot diseases of poplars caused by *Septoria musiva* and *S. populicola*. Phytopathology 31:241-254. A classic paper on the topic.

Thordal-Christensen H. (2003) Fresh insights into processes of nonhost resistance. Curr Opin Plant Biol 6:351-357.

Tomescu R, Nef L.(2007) Leaf eating insect damage on different poplar clones and sites. Ann For Sci 64:99-108.

Tuskan GA, Difazio S, Jansson S, Bohlmann J, Grigoriev I, Hellsten U, et al.(2006) The genome of black cottonwood, *Populus trichocarpa* (Torr. & Gray). Science 313: 1596-1604. A description of the sequencing of the black cottonwood genome and some insight into how such work has now heralded a new era in understanding trees.

Verani S, Sperandio G, Picchio R, Spinelli R, Picchi G. (2008) Field handbook – *Poplar* harvesting. International Poplar Commission Working Paper IPC/8, Rome, Italy. Forestry Department, Food and Agriculture Organization of the United Nations.

Wang J, Van Der Kamp B. (1992) Resistance, tolerance, and yield of western black cottonwood infected by *Melampsora* rust. Can J For Res 22:183-192.

Ward KT, Ostry ME. (1994) Morphological and molecular genetic variation among isolates of *Septoria musiva*. Phytopathol 84:1145.

Ward KT, Ostry ME. (2005) Variation in *Septoria musiva* and implications for disease resistance screening of poplars. Plant Dis 89:1077-1082.

Waterman AM. (1946) Canker of hybrid poplar clones in the United States, caused by *Septoria musiva*. Phytopathology 35:148-156. A historically important paper in understanding fungal disease.

Waterman AM, Aldrich KF. (1952) Surface sterilization of poplar cuttings. Plant Dis Rep 36:203-207.

Waterman AM. (1954a) Surface sterilization of hybrid poplar cuttings. Forest Research Notes, 32.

Waterman AM. (1954b) *Septoria* canker of poplars in the United States. United States Department of Agriculture, Circular No. 947. A historically important paper in understanding fungal disease.

Weber JC, Stettler RF. (1981) Isoenzyme variation among ten populations of *Populus trichocarpa* Torr. et Gray in the Pacific Northwest. Silv Genet 30:82-87. Used clones from British Columbia.

Weber JC, Stettler RF, Heilman PE. (1985) Genetic variation and productivity of *Populus trichocarpa* and its hybrids. I. Morphology and phenology of 50 native clones. Can J For Res 15:376-383. Used clones from British Columbia.

Weiland JE, Stanosz JC, Stanosz GR. (2003) Prediction of long-term canker disease damage from the responses of juvenile poplar clones to inoculation with *Septoria musiva*. Plant Dis 87:1507-1514.

Weiland JE, Stanosz GR. (2006) Cultural and PCR-based detection of *Septoria musiva* in inoculated hybrid poplar stems. Forest Pathol 36:198-208.

Weiland JE, Stanosz GR. (2007) The histology of hybrid poplar clones inoculated with *Septoria musiva*. Plant Dis 91:1524-1530. A detailed description of the microscopic appearances of disease.

Woodbury PB, Laurence JA, Hudler GW. (1994) Chronic ozone exposure increases the susceptibility of hybrid *Populus* to disease caused by *Septoria musiva*. Environ Pollution 86:109-114.

Working Paper IPC/9-4a, Rome, Italy. Forestry Department, Food and Agriculture Organization of the United Nations.

Yang D, Plante F, Bernier L, Piche Y, Dessureault M, Laflamme G, Ouellette GB. (1993) Evaluation of a fungal antagonist, *Phaeotheca dimorphospora*, for biological control of tree diseases. Can J Bot 71:426-433.

Yang D, Bernier L, Dessureault M. (1994) Biological control of *Septoria* leaf spot of poplar by *Phaeotheca dimorphospora*. Plant Dis 78:821-825.

Zalasky H. (1964) The histopathology of *Macrophoma tumefaciens* infections in black poplar. Can J Bot 42:385-391.

Zalasky H, Fenn OK, Lindquist CH. (1968) Reactions of poplars to infection by *Septoria musiva* and *Diplodia tumefaciens* and to injury by frost in Manitoba and Saskatchewan. Plant Dis Rep 52:829-833.

Zalasky H. (1978) Stem and leaf spot infections caused by *Septoria musiva* and *S. populicola* on poplar seedlings. Phytoprotection 59:43-50. A Canadian paper with detail on the pathogenesis of disease which also provides some data re: canker caused by *S. populicola*.

Zhang P, Dreher K, Karthikeyan A, Chi A, Pujar A, Caspi R, Karp P, Kirkup V, Latendresse M, Lee C, Mueller LA, Mueller R, Rhee SY. (2010) Creation of a genome-wide metabolic pathway database for *Populus trichocarpa* using a new approach for reconstruction and curation of metabolic pathways for plants. Plant Physiol 153:1479-1491.