Eur. J. For. Path. 25 (1995) 241–252

ISSN 0300–1237

Northeastern Forest Experiment Station, USDA Forest Service, Durham, New Hampshire, USA

Response of maple sapwood to injury and infection

By W. C. SHORTLE, K. T. SMITH, K. R. DUDZIK and S. PARKER

Summary

In sapwood challenge experiments in *Acer rubrum*, columns of discolouration initiated by wounding and inoculation with pioneer fungi (*Cephalosporium* sp., *Phialophora* sp.) were similar in size to untreated wounds. Inoculation with decay fungi (*Pleurotus ostreatus, Trametes versicolor*) produced larger columns of wound-initiated discolouration. The removal of bark around a bore wound caused a significantly larger column to form compared to the sum of the columns inititiated by separate wounds. Stage-I discoloured wood, not associated with obviously rotted wood, had concentrations of mobile cations and soluble phenols similar to sapwood. Stage-II discoloured wood, spatially associated with rotted wood, was frequently bounded by a chemically distinct boundary layer and the discoloured wood contained significantly greater concentrations of mobile cations and soluble phenols than stage-I discoloured wood.

1 Introduction

The wounding of sapwood initiates a cascade of changes that involve the defense system of the tree and the exploitation of opportunities by an assemblage of micro-organisms. In red maple (*Acer rubrum* L.), and many other tree species, wounding initiates the process that discolours sapwood. Heartwood, the result of normal aging in many tree species, is not formed in red maple. Central columns of discoloured sapwood in red maple result from wound-initiated discolouration (SHIGO 1984). Although wounding results in the aeration and desiccation of sapwood, wood discolouration is due to enzymatic oxidation of wood compounds rather than changes in water distribution (SHIGO 1984). Eventually, the structural breakdown or decay of wood in living trees occurs in wood that has previously discoloured.

In recent decades, two conceptual models have been developed to describe this process of wound-initiated transformation. Both models refer to the limits of this transformation as the result of the process of compartmentalization. Compartmentalization is a boundary-setting process that tends to minimize the amount of sapwood that is transformed by wound-initiated discolouration and decay (SHIGO 1984).

The first of the two conceptual models is a three-stage temporal model (SHIGO and HILLIS 1973). In the temporal model, the concept of organismal succession was a key element. Wounding initiates a series of host responses (stage I) that is followed by the infection and spread of pioneer micro-organisms that interact with the host tree (stage II), and the process is eventually completed by the decomposition of wood by decay micro-organisms (stage III). In this model, the development and spread of discoloured wood associated with a wound occurs as part of both stage I and stage II.

The second of the two conceptual models is a spatial model that involves the formation of distinct, physical boundaries that localize or 'wall-off' infections and tend to limit their spread (SHIGO 1984; SHORTLE and SMITH 1990). The concept of geometric orientation of pre-existing and newly induced physical and chemical boundaries is a key element in the second model. Wound-initiated discolouration develops within the boundaries defined by the compartmentalization process.

Linkage of the temporal and spatial models may provide a better understanding of the spread of infection that causes the loss of wood quality in living trees. Experiments were conducted for a series of three objectives using red maple trees. The first objective was to compare the effects of inoculation with decay fungi and 'pioneer' non-decay fungi on the size of columns of discoloured wood initiated by wounding. The second objective was to determine the effect of the loss of living phloem and the length of living rays on the size of columns of discolouration. The third objective was to compare the chemical characteristics of stage-I and stage-II discolouration.

2 Materials and methods

2.1 Experimental trees

All experiments were conducted on dominant and codominant red maple trees from mixed northern hardwood stands in Maine and New Hampshire, USA.

2.2 *In situ* challenges of sapwood

The healthy sapwood of living red maple trees was challenged by decayed wood or fungi associated with wood discolouration and decay in three separate experiments. In sapwood challenge 1, freshly wounded recipient trees were inoculated with wood previously altered by the discolouration and decay process in donor trees. Donor trees were wounded 3 years prior to the wounding and inoculation of recipient trees to initiate wood discolouration and decay. Wounds in donor trees consisted of a single radial hole drilled at 1.4 m above ground-line with a 0.5-cm-drillbit to a depth of 2.5 cm. Wood discolouration and decay proceeded in donor trees without further treatment. To obtain inoculum for recipient trees, an increment borer was used to extract cores from columns of wound-initiated discolouration and decay produced in donor trees. An aligned series of cores was taken, parallel to and above and below the previously drilled wound in the donor tree. The cores of donor trees were cut to yield 2-cm-long segments of unaltered sapwood (SW), discoloured wood (DW), and rotted wood (RW).

Five recipient trees (10–15 cm dbh) were wounded in September and immediately inoculated with core segments from donor trees. Each tree received six wounds 1 m above ground level and again at 2 m above ground level. Wounds were evenly spaced around the stem circumference and consisted of radial holes drilled to a depth of 2.5 cm with 0.5-cm-diameter bit. The wounds in the upper whorl were not vertically aligned with the wounds in the lower whorl. In each whorl of six wounds, holes were inoculated with donated segments of SW, DW, RW, and one hole was left uninoculated. The remaining two holes received additional segments of SW, DW, or RW.

Prior to harvest, one of the recipient trees died for unknown reasons. 1 year after inoculation, The four remaining recipient trees were felled and stem disks sawn at 4-cm intervals. The volume of wound-initiated discolouration and decay associated with the various treatments was calculated from serial stem disks. Mean values for the volume of discoloured and decayed wood associated with each inoculum treatment were compared to volumes of discolouration and decay using the protected least-significant difference, calculated for unequally replicated treatments ($p \leqslant 0.01$).

In sapwood-challenge 2, fresh drill wounds of recipient trees were treated with segments of wood and with pure cultures of fungi associated with the discolouration and decay process. A total of 10 recipient trees (10–15 cm dbh) were wounded in May at 1 m above ground-level, as described above. These wounds were left open or inoculated with increment core segments that contained SW, DW, and RW obtained from donor trees, as in sapwood challenge 1. Donor trees were felled and bolts containing the increment borer holes were

returned to the laboratory. Under aseptic conditions, the bolts were split radially along the length of the borer holes. Small wood chips were aseptically gouged from between the borer holes and plated on malt-yeast-extract agar (SHIGO and SHARON 1968). Fungal cultures were purified according to standard methods. The most commonly occuring hymenomycetous decay fungi and phialosporous hyphomycetes ('pioneer fungi' in SHIGO and HILLIS 1973) were selected as additional inoculants for sapwood-challenge 2. Autoclaved 2-cm-long segments of red maple sapwood were placed on cultures of single isolates of the decay fungi *Trametes versicolor* (L.:Fr.) Pilat and *Pleurotus ostreatus* (Jacq.:Fr.) Kummer and of pioneers in the genera *Phialophora* Medlar and *Cephalosporium* Corda. Additional autoclaved core segments were placed on sterile, un-inoculated medium. In June, after the core segments were covered by mycelium (except for the un-inoculated controls), the segments were taken to the field. An upper whorl of wounds was made at 2 m above ground level in the same trees wounded in the previous month, and the drill wounds inoculated with the colonized core segments or sterile control segments. Prior to harvest, one of the recipient trees died for unknown reasons.

After 2 years incubation, the nine remaining recipient trees of sapwood-challenge 2 were felled and stem disks were sawn at 4-cm intervals. The total length of discolouration associated with each wound and the mean width at 2–4 cm above and below the wound were recorded. Mean values for each inoculum treatment were compared to un-inoculated controls using the protected least-significant difference, calculated for unequally replicated treatments ($p \leqslant 0.01$).

In sapwood-challenge 3, 15 red maple trees (18–25 cm dbh) were wounded and inoculated in August with sapwood core segments colonized by pure cultures of decay and pioneer fungi. The wounds and inoculations were made as described for the upper whorl of wounds in sapwood-challenge 2. Inoculum consisted of single red-maple isolates of *T. versicolor, P. ostreatus, Phialophora* sp., as used for challenge 2, as well as single red-maple isolates of pioneers in the genera *Graphium* Corda, *Fusarium* Link, a single beech isolate of the decay fungus *Phellinus igniarius* (L.:Fr.) Quel. and the oak isolates *Daedalea quercina* (L.:Fr.) Fr. and *Laetiporus sulphureus* (Bull.:Fr) Murrill. Four trees were harvested each September after 1, 2, and 3 years incubation, respectively. The width of each column of discolouration at 2–4 cm above and below each wound was recorded. Mean values for each inoculum treatment were compared to un-inoculated controls using the protected least-significant difference, calculated for unequally replicated treatments ($p \leqslant 0.01$)

2.3 Effect of bark removal on area of wound-initiated discolouration

Five red-maple trees (10–15 cm dbh) were wounded to assess the effect of bark removal on the transverse area of columns of wound-initiated discolouration. Each tree received three different wounds in a single whorl at 1 m above ground level. Wounds consisted of: 1. removal of a strip of bark 2.5 cm wide (measured along the stem circumference) × 10 cm long (measured along the vertical axis of the stem); 2. a bore hole 1.3 cm in diameter drilled to a radial depth of 3 cm; and 3. A combination of wounds 1 and 2 with the bore hole centered in the area of removed bark. After 2 years, the trees were felled and a 4-cm-thick disk was sawn from immediately above the drill wounds. A clear plastic sheet was placed on the upper transverse surface of the sawn disk from each tree. The discoloured and decayed area associated with each wound was traced on the plastic sheet and measured with a planimeter.

The areas of discolouration associated with the separate bark removal and bore hole wounds were summed and compared to the areas of discolouration associated with the combined bore-hole and bark-removal wounds by an F-test ($n = 5$, $p \leqslant 0.05$).

2.4 Effect of phloem contact and living ray length on the chemistry of column-boundary layers

Five red-maple trees (12–20 cm dbh) that each contained a previously formed central column of wound-initiated discolouration were selected for wounding. Trees were wounded by drilling four evenly spaced holes in each of three whorls at 1.0, 1.5, and 2.0 m above ground level. Each wound was 1.4 cm in diameter and 5 cm in length. Within each whorl, wounds were angled to be $>45°$, $45°$, $<45°$, and $0°$ with respect to the rays. After 2 years, the trees were harvested and the bolts containing the columns of discolouration initiated by the angled wounds were sawn into 4-cm-thick disks. In the stem disks, the outer edge of each angled column of discolouration was identified as the margin or boundary of the column that faced the phloem and cambium. The inner edge of each angled column was the margin or boundary that faced the central column of discolouration. For each angled wound, the ray length was measured as the distance along the stem radius from the inner edge to the central column of discolouration.

For chemical analysis, blocks of 1 × 2 × 4 cm (radial: tangential: longitudinal) containing discolouration initiated by the angled wounds were split and freeze-dried. Shavings containing outer and inner boundary layers, sapwood, and discoloured wood were removed from the blocks with a wood plane. The shavings from each type of wood were milled separately to pass through a 250 μm sieve. Water-soluble dry matter and total water-soluble phenols were determined from triplicate 0.5 g samples of milled wood, as described previously (SHORTLE 1979). Following inspection of the plotted relationships of wood chemistry and sample position, the concentrations of total soluble matter and total soluble phenols in the inner boundary were regressed against ray length.

2.5 Chemical differentiation of stage-I and stage-II discoloured wood

Two sets of six red-maple trees (30–45 cm dbh) were compared. Each tree in the first set contained a central column of discolouration, initiated by natural wounding prior to this experiment (PARKER et al. 1994: Fig. 1). No decay was associated with these columns and the discolouration was evaluated as being of type I, due to the host response to wounding. Approximately 10 years prior to this experiment, trees in the second set had been wounded by drilling or by drilling and the removal of bark (PARKER et al. 1994: Fig. 2). Decayed wood was clearly associated with the columns, and the discolouration was evaluated as being of type II, due to the interaction between the wounded host tree and micro-organisms. Trees were felled and 5-cm-thick disks were sawn from the stems of trees in both sets. Disks sawn from the first set of trees contained sapwood, stage-I discoloured wood, and the boundary between sapwood and stage-I discoloured wood. Disks sawn from the second set of trees contained sapwood, stage-II discoloured wood, stage-III discoloured wood (= rotted wood), and the boundary between sapwood and stage-II discoloured wood. Wood samples were taken as drill shavings, removed using a 3-mm-diameter cobalt bit bored 2 cm into the transverse surface of the air-dried disks. For each tree and wood type, the drill shavings from 9 holes were pooled.

Chemical extracts were prepared by placing 50-mg samples of each wood type in a 5-ml vial followed by an addition of 3 ml of deionized, distilled water. After incubation at 22 ± 1°C for 1.5 h, the electrical resistance of the slurry was measured using a model OZ—67 Shigometer (Osmose Wood Preserving Co., Buffalo, NY, USA) and a Delmhorst No. 2 E detection electrode fitted with 20-mm stainless-steel contact pins at a spacing of 11 mm, and held in a No. 552/A-106 retainer (Delmhorst Instrument Co., Towaco, NY, USA). The pins were immersed so that the tips of the pins rested on the bottom of the sample vial. Electrical resistance was measured and converted to the equivalent concentration of K^+ (mmol/l) using a standard curve of electrical resistance vs. concentration of KCl. Following

measurement of electrical resistance, sample shurries were incubated overnight in order to stabilize the absorbance of UV light. After 18 h, 0.5 ml of the sample liquid was pipetted into a fresh vial and diluted with 2.0 ml of distilled, deionized water. Absorbance of the diluted solution was measured at 300 nm in a spectrophotometer. Absorbance was converted to the equivalent concentration of the soluble phenol gallic acid (mmol/l) using a standard curve of absorbance vs. gallic acid concentration. The mean and confidence interval ($p \leqslant 0.05$) for each set of trees and wood type were calculated and compared.

3 Results

3.1 *In situ* sapwood challenges

The effect of inoculation on the size of columns of wound-initiated discolouration and decay in red maple was tested in three experiments. These three experiments challenged freshly drilled sapwood of living red-maple trees with wood altered by the decay process and with pure cultures of fungi associated with discolouration and decay. In sapwood-challenge 1, after 1 year of incubation, greater volumes of discolouration resulted from wounds treated with rotted wood than from untreated control wounds ($p \leqslant 0.01$; Table 1). There were no significant differences in the volume of discolouration between untreated control wounds and wounds treated with sapwood or discoloured wood.

Similarly, after 2 years of incubation in sapwood-challenge 2, columns of discolouration were longer and wider following the treatment of wounds with rotted wood compared to untreated controls. Treatment of wounds with sapwood or discoloured wood had no significant effect on column length or width. The inoculation of drill wounds with pure cultures of the decay fungi *T. versicolor* and *P. ostreatus* resulted in significantly longer and wider columns of discolouration than columns produced by untreated control wounds. The

Table 1. **Mean measurements (and number of observations) of decay columns in red maple following the challenge *in situ* of wounded sapwood**

Sapwood challenge:	1		2	3		
Measurement variable:	Volume (cm^3)		Length (cm), width (mm)	Width (mm)[1]		
Incubation period (years):	1		2	1	2	3
Treatment	Wood inoculum		Culture inoculum			
Rotted wood	100 (10)**	13.6, 8.0 (11)**				
Discoloured wood	32 (15)	9.1, 6.4 (20)				
Untreated sapwood	44 (15)	9.1, 6.0 (13)				
Control (no treatment)	42 (16)	9.6, 6.2 (9)	9.7, 6.4 (7)	5.5	7.2	8.0
Sterile sapwood			12.0, 6.9 (9)	6.2	8.0	10.7
Phialophora sp.			11.6, 7.4 (9)	6.2	7.8	9.8
Cephalosporium sp.			12.2, 7.1 (9)			
Graphium sp.				5.8	7.7	9.3
Fusarium sp.				5.8	7.3	9.5
Trametes versicolor			24.9, 11.0 (9)**	9.5**	9.5	10.3
Pleurotus ostreatus			22.4, 9.0 (8)**	7.0	7.7	8.5
Daedalea quercina				5.5	7.2	9.7
Laetiporus sulphureus				4.8	7.5	10.2

[1] Each measurement value for sapwood-challenge 3 is the mean of four observations.
** All statistical comparisons are within table columns; signficantly different from control at $p \leqslant 0.01$

size of columns of discolouration was not affected by the inoculation of wounds with sterile sapwood or pioneer fungi (Table 1).

In sapwood-challenge 3, after 1 year of incubation, the inoculation of drill wounds with *T. versicolor* resulted in wider columns of discolouration than untreated control wounds (Table 1). After 2 and 3 years of incubation, the mean width of discolouration produced by all treatments were not significantly different from untreated controls.

3.2 Effect of bark removal on area of wound-initiated discolouration

The separate bore-hole and bark-removal wounds produced well-defined columns of wound-initiated discolouration (Fig. 1). The combined bore-hole and bark-removal wounds produced somewhat amorphous columns with spreading, uneven boundaries. The mean area of discolouration of the combined bore-hole and bark-removal wounds (29 cm^2) was significantly greater ($p \leq 0.05$) than the mean sum of the areas of discolouration associated with separate bore-hole and bark-removal wounds (11 cm^2).

3.3 Effect of phloem contact and living ray length on the chemistry of column-boundary layers

Angled wounds in trees that already contained compartmentalized central columns of discolouration resulted in the formation of new columns of discoloration in which the lateral boundaries of the column (following the length of the bore hole) were of two types,

Fig. 1. Transverse view of columns of wound-initiated discolouration in a stem disk of red maple. Clockwise from the lower right, wounds consisted of a single bore hole, the removal of bark, and the combination of both bark removal and a bore hole

inside and outside (Fig. 2). The outside boundary was connected to the phloem by living ray cells. The inner boundary was connected to living rays that were cut off from direct connection with the phloem by the pre-existing central column of discolouration.

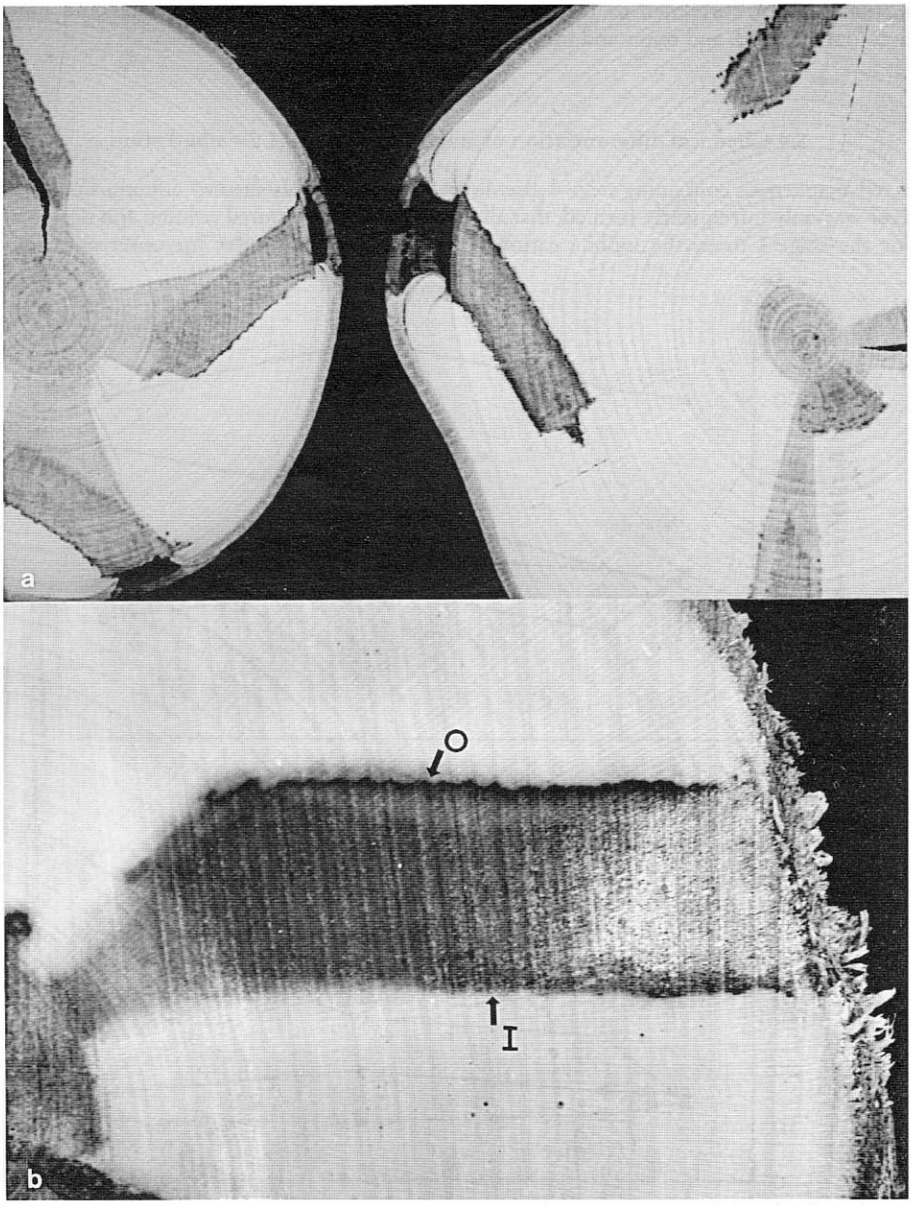

Fig. 2. Transverse view of columns of wound-initiated discolouration in stem disks of red maple: (a). Wounds were bore holes, angled away from the stem radius; (b). Such wounds produced columns with an outer boundary (arrow marked 'O') connected to the phloem by rays and an inner boundary (arrow marked 'I') with no direct connection to the phloem. The outer boundary consists of a visibly and chemically discrete column-boundary layer. The inner boundary is an undifferentiated interface between discoloured wood and sapwood

The outside boundaries, connected to the phloem by living rays, had the highest concentrations of soluble dry matter and soluble phenol (Fig. 3). In the inside boundary, the concentration of soluble dry matter and total phenol was highly correlated with total ray length ($r^2 = 0.98$ and 0.90, respectively). The concentration of soluble dry matter and soluble phenol was not correlated to total ray length in the outer boundary, outer and inner discoloured wood, and outer and inner sapwood.

3.4 Chemical differentiation of stage-I and stage-II discolouration

In red-maple trees containing stage-I discoloured wood, a visibly distinct column-boundary layer was only infrequently formed, if at all, between the discoloured column and sapwood. For the stage-I trees, the mobile cation concentration was similar for outer and inner

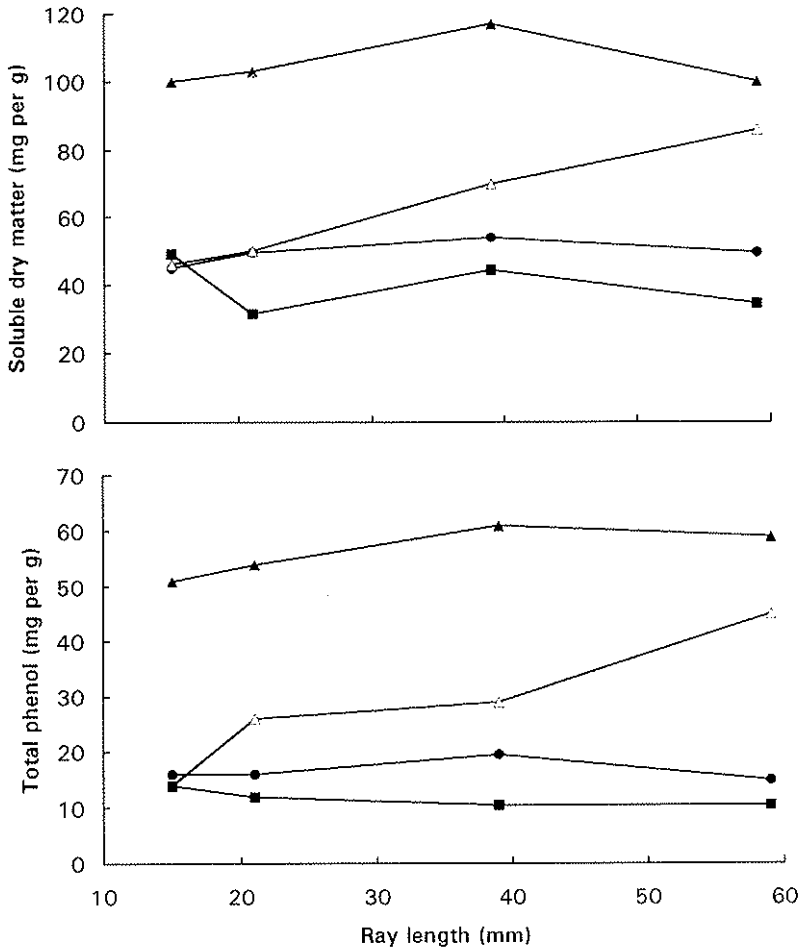

Fig. 3. The effect of ray length on the concentration of soluble dry matter (top) and soluble phenol (bottom) in sapwood (black circle), discoloured wood (black rectangle), outer-column-boundary layer (black triangle), and inner-column boundary (white triangle)

sapwood, stage-I discoloured wood, and the boundary between inner sapwood and stage-I discoloured wood (Table 2). Similarly, the concentration of soluble phenol in the sapwood, stage-I discoloured wood, and the boundary were not significantly different. In trees containing stage-II discoloured wood, a visibly distinct column-boundary layer formed between inner sapwood and stage-II discoloured wood. Outer and inner sapwood contained significantly lower concentrations of mobile cations and soluble phenols than the other types of wood tested from trees with stage-II discolouration (Table 2).

4 Discussion

Two apparently separate sets of processes occur in the development of wound-initiated discolouration and decay. One set of processes results in the spatial positioning of the boundaries of compartmentalization. The second set of processes consists of the biological and chemical changes within the boundaries.

The inoculation of drill wounds with rotted wood in sapwood-challenge 1 and 2 caused the boundaries of compartmentalization to form at a greater distance from the wound, resulting in larger columns of wound-initiated discolouration. The inoculation of wounds with discoloured wood or sapwood had no measurable effect compared to control wounds that received no treatment. This was interpreted as evidence that the position of the compartmentalization boundaries is due to the activity of decay fungi and not primarily to the activity of pioneers that may also be present in discoloured wood. This was also taken as evidence that the position of the boundaries is not solely an attempt by the tree to maintain sapwood moisture, as has been suggested (BODDY and RAYNER 1983). When pure-culture inoculum was used in sapwood-challenge 2, the wood-decay fungi *T. versicolor* and *P. ostreatus* caused longer and wider decay columns to form compared with controls. Wood-decay fungi are not the only micro-organisms that cause the formation of enlarged columns of wound-initiated discolouration. For example, the inoculation of *Acer saccharum* Marsh with the agressive vascular pathogen *Ceratocystis virescens*, causal agent of sapstreak disease,

Table 2. **Mean concentration of mobile cations and soluble phenols in wood associated with discolouration and decay in red maple**

Wood type	Mobile cations (K^+ equiv, mmol/l)		Soluble phenol (gallic acid equiv, mmol/l)	
	Stage-I trees	Stage-II trees	Stage-I trees	Stage-II trees
Outer sapwood	0.6 ± 0.2[1]	0.6 ± 0.2	0.33 ± 0.04	0.50 ± 0.10
Inner sapwood	0.5 ± 0.2	0.5 ± 0.2	0.35 ± 0.05	0.45 ± 0.15
Stage-I discoloured wood	0.6 ± 0.2		0.37 ± 0.09	
Stage-II discoloured wood		5.2 ± 1.5		1.22 ± 1.33
Inner sapwood/stage-1 boundary	0.6 ± 0.3		0.28 ± 0.05	
Inner sapwood/stage-2 boundary layer		2.7 ± 1.1		1.40 ± 0.45
Rotted wood		7.0 ± 1.6		

[1] Confidence intervals ($p \leq 0.05$) were calculated from the standard error of the mean and the t-distribution ($n = 6$)

has caused the formation of extensive columns of wound-initiated discolouration (SMITH and HOUSTON 1994).

Inoculation of wounds with the pioneer fungi *Phialophora* sp. or *Cephalosporium* sp. produced columns of discolouration not significantly different in length or width from controls. The pioneer fungi may well have an ecological role in influencing the pattern of subsequent colonization in wound-altered wood. Although a rigidly deterministic succession is unlikely, the pioneers may influence the colonization and exploitation of columns of discolouration and decay through both competitive exclusion and detoxification of protective chemicals produced by defensive processes in the tree (SMITH et al. 1981).

After 1 year of incubation in sapwood-challenge 3, columns of discolouration and decay associated with wounds inoculated with *T. versicolor* were significantly wider than columns associated with control wounds. No effect on column width was attributable to the other decay or pioneer fungi. After 2 and 3 years of incubation, none of the treatments resulted in a column width significantly different from control. As no attempt was made at fungal recovery, it is not known how infection of wounds with native inoculum affects the width of columns associated with control wounds. The increased energy reserves in the larger trees used in sapwood-challenge 3 may have enabled the tree to resist the spread of infection to a greater degree than in the smaller trees used in sapwood-challenges 1 and 2. Furthermore, the width of columns may not be as sensitive a measure of decay-column development as column volume or length and width. Column volume as measured in sapwood-challenge 1 is most desirable as a sensitive indicator of the effectiveness of compartmentalization, but is laborious to accomplish.

The combined bore-hole wound plus bark removal initiated significantly larger columns of discolouration after 2 years than the additive effect of the wounds made separately. The removal of bark directly kills sapwood cells by desiccation. More importantly, removal of bark also removes the phloem that supplies sapwood with carbohydrates and amino acids for respiration and biosynthesis. The large columns of discolouration associated with the combined wounds are here considered to be due to the inability of the tree to set effective boundaries to the spread of damage caused by the bore hole.

The angled wound experiment emphasized the importance of the supply of energy and biosynthetic materials. The greater amounts of soluble dry matter and soluble phenol in the outer boundary are considered to be a result of the comparatively greater access to this material through the rays in direct contact with the phloem. The concentration of soluble dry matter and phenol in the inner boundary was highly correlated with ray length. It is suggested that the soluble dry matter and phenol in the column boundaries was mobilized or synthesized from reserves in the ray cells, and it is expected that longer rays would be able to supply an inner boundary with greater amounts of energy and biosynthetic intermediates, resulting in higher concentrations of soluble dry matter and phenol. The lack of correlation of concentration in the outer boundary with ray length may be due to the direct access to the phloem, making the amount stored in the rays of less significance. The lack of correlation of sapwood concentration with ray length is reasonable in that sapwood that is not responding to injury and infection would not be importing appreciably high amounts of stored reserves from rays. The lack of correlation of concentration in the discoloured wood is predictable in that the discoloured wood, of stage-I or stage-II, is cut off from the symplast and the energy system of the tree.

Previous research (SHORTLE and SMITH 1990) described a chemically distinct column-boundary layer that partially formed between a column of discolouration and sapwood. In the results reported here, there was no differentiated layer at the interface between sapwood and stage-I discoloured wood, the latter being the product of host response to wounding. A visibly distinct boundary layer did form between sapwood and stage-II discoloured wood, the latter being the product of interaction between the host tree and microorganisms. We suggest that the distinct boundary layer is formed by the tree only in response to infection.

Conclusion

The spatial position of compartmentalization boundaries was due to the interaction between the wounded red-maple trees and the decay fungi. Inoculation of wounds with pioneer micro-organisms did not affect the size of compartmentalized columns of wound-initiated discolouration. The effectiveness of the tree in limiting the size of columns was related to access to living phloem and ray cells. Although visually similar, stage-I discolouration (interpreted as the result of tree response to wounding) and stage-II discolouration (interpreted as the result of interaction with micro-organisms) are chemically distinct. Useful interpretation of the discolouration and decay process requires clarification of the separate and interactive roles of micro-organisms and tree response following injury and infection.

Résumé

Réponse de l'aubier de l'Erable rouge aux blessures et aux infections

Dans des essais sur *Acer rubrum*, les colonnes de coloration initiées dans l'aubier par des blessures inoculées avec des champignons pionniers, avaient des dimensions semblables à celles produites par des blessures non inoculées. Après inoculation par des champignons d'altération, les colonnes de coloration de blessure étaient plus grandes. L'enlèvement de l'écorce autour de la blessure provoquait une colonne significativement plus grande comparée à la somme des colonnes initiées par des blessures séparées. L'aubier et le stade I de coloration du bois, non associé à une pourriture évidente, avaient des concentrations semblables en cations mobiles et en phénols solubles. Le stade II de coloration du bois, spatialement associé au bois pourri, était fréquemment bordé par une limite chimiquement distincte et le bois coloré avait des concentrations significativement plus grandes en cations mobiles et en phénols solubles.

Zusammenfassung

Reaktion von Ahorn-Splintholz auf Verletzung und Infektion

In Experimenten, bei denen Splintholz von *Acer rubrum* verletzt bzw. mit Pilzen aus der Gruppe der Primärbesiedler (*Cephalosporium* sp., *Fusarium* sp., *Graphium* sp., *Phialophora* sp.) inokuliert wurde, entwickelten sich unabhängig von der Behandlung etwa gleich große Bereiche verfärbten Holzes. Die Inokulation von Fäuleerregern (*Pleurotus ostreatus, Trametes versicolor*) führte dazu, daß sich von der Verletzungsstelle aus größere Holzbereiche verfärbten. Wenn die Rinde um das Bohrloch entfernt wurde, war der verfärbte Holzbereich größer als die Summe der von separaten Verletzungen (Bohrloch und Rindenverletzung) ausgehenden Verfärbungen. Verfärbtes Holz der Stufe I, das nicht offensichtlich mit einer Fäule assoziiert war, enthielt ähnliche Konzentrationen an mobilen Kationen und löslichen Phenolen wie der normale Splint. Holz der Verfärbungsstufe II, das räumlich mit verfaultem Holz assoziiert war, war häufig von einer chemisch unterscheidbaren Zone umgeben, und das verfärbte Holz enthielt signifikant höhere Konzentrationen an mobilen Kationen und löslichen Phenolen.

References

BODDY, L.; RAYNER, A. D. M., 1983: Origins of decay in living deciduous trees: the role of moisture content and re-appraisal of the expanded concept of tree decay. New Phytol. **94**, 623–641.

PARKER, S.; SHORTLE, W. C.; SMITH, K. T., 1994: Identification of gram-positive bacteria isolated from initial stages of wound-initiated discoloration of red maple. Eur. J. For. Path. **24**, 48–54.

SHIGO, A. L., 1984: Compartmentalization: a conceptual framework for understanding how trees grow and defend themselves. Ann. Rev. Phytopathol. **22**, 189–214.

–; HILLIS, W. E., 1973: Heartwood, discolored wood, and microorganisms in living trees. Ann. Rev. Phytopathol. **11**, 197–222.

–; SHARON, E. M., 1968: Discoloration and decay in hardwoods following inoculation with *Hymenomycetes*. Phytopathology **58**, 1493–1498.

SHORTLE, W. C., 1979: Compartmentalization of decay in red maple and hybrid poplar trees. Phytopathology 69, 410–413.
–; SMITH, K. T., 1990: Decay column boundary layer formation in maple. Biodeterioration Research 3, 377–389.
SMITH, K. T.; BLANCHARD, R. O.; SHORTLE, W. C., 1981: Postulated mechanism of biological control of decay fungi in red maple wounds treated with *Trichoderma harzianum*. Phytopathology 71, 496–498.
–; HOUSTON, D. R., 1994: Metal concentrations in wood of sugar maple with sapstreak disease. Can. J. For. Res. 24, 185–188.

Authors' address: Dr. Walter C. SHORTLE, Dr. Kevin T. SMITH (for correspondence), Kenneth R. DUDZIK and Sharon PARKER, USDA Forest Service, PO Box 640, Durham, NH 03824–0640, USA

Received: 22.11.94; accepted: 20.4.1995