Ethanol and acetone from Douglas-fir roots stressed by Phellinus sulphurascens infection: Implications for detecting diseased trees and for beetle host selection

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## A1. Results

## A1.1. Sites 1, 2 root headspace acetaldehyde, methanol, and propanol

Acetaldehyde concentrations in the root phloem and xylem were both dependent on tree condition, or month, but not their interaction (Appendix A, Table A1). The $2.2 \mu \mathrm{~g} \mathrm{~g}^{-1}$ fresh mass concentration in phloem from diseased roots was 3.6 (95\% CI 2.0, 6.8) times greater than in healthy trees. September phloem contained $3.6 \mu \mathrm{~g} \mathrm{~g}^{-1}$ fresh mass acetaldehyde, which was comparable to the $3.56 \mu \mathrm{~g} \mathrm{~g}^{-1}$ fresh mass in November roots ( $\mathrm{t}_{46.3}=0.18, P=0.858$ ), but both were greater than the $0.12 \mu \mathrm{~g} \mathrm{~g}^{-1}$ fresh mass in May roots ( $\mathrm{t}_{36.8}=11.94, P<0.001$ and $\mathrm{t}_{41.9}=$ $11.84, P<0.001$, respectively). In xylem, the $15.0 \mu \mathrm{~g} \mathrm{~g}^{-1}$ fresh mass of acetaldehyde in diseased tree roots was 3.3 ( $95 \%$ CI 2.1, 5.3) times greater than in healthy trees $\left(\mathrm{t}_{42.5}=5.15, P<0.001\right.$ ). Tree root xylem in September contained $10.3 \mathrm{\mu g} \mathrm{~g}^{-1}$ fresh mass acetaldehyde, similar to the 8.7 $\mu \mathrm{g} \mathrm{g}^{-1}$ fresh mass in November root xylem ( $\mathrm{t} 40.7=1.26, P=0.214$ ). The September xylem concentration was 1.7 ( $95 \%$ CI 1.1, 2.5) times higher than in May roots ( $\mathrm{t}_{48.5}=2.56, P<0.014$ ). The November and May root xylem concentrations were not different ( $\mathrm{t}_{44.8}=1.79, P<0.081$ ).

Methanol in root phloem was not different between diseased and healthy trees (10.2 vs. 8.7 $\mu \mathrm{g} \mathrm{g}{ }^{-1}$ fresh mass), among months, or their two way interaction (Appendix A, Table A1).

However, xylem methanol did exhibit a tree condition by month interaction (Appendix A, Table A1). In September the $10.6{\mu g g^{-1}}^{\text {fresh mass xylem concentration in diseased trees was higher }}$ $(\mathrm{t} 4.2=2.87, P=0.042)$ than the $6.2 \mu \mathrm{~g} \mathrm{~g}$ - fresh mass in healthy trees. A similar difference occurred in May when diseased tree xylem contained $11.1 \mu \mathrm{~g} \mathrm{~g}{ }^{-1}$ fresh mass of methanol compared to $7.1 \mu g ~ g^{-1}$ fresh mass in healthy trees ( $\mathrm{t}_{5.98}=3.08, P=0.022$ ). However, in November there was no difference ( $\mathrm{t} 1.0=1.68, P=0.342$ ) in methanol concentrations between the diseased and healthy trees ( 9.9 vs. $8.3 \mu \mathrm{~g} \mathrm{~g}^{-1}$ fresh mass, respectively).

Propanol concentrations in the phloem were dependent on tree condition only (Appendix A, Table A1). The $0.30 \mu \mathrm{~g} \mathrm{~g}^{-1}$ fresh mass in diseased tree roots was 3.8 ( $95 \%$ CI 1.7, 8.8) times greater than in healthy tree phloem ( $\mathrm{t}_{35}=3.26, P=0.003$ ). In xylem, the propanol concentrations were dependent on tree condition, or month, but not their two-way interaction (Appendix A, Table A1). The $0.6 \mu \mathrm{~g} \mathrm{~g}$-1 fresh mass of propanol in xylem of diseased tree roots was 2.83 ( $95 \%$ CI 1.4, 5.8) times greater than in healthy tree roots $\left(\mathrm{t}_{38.3}=2.94, P=0.006\right)$. The May root xylem propanol concentration of $0.6 \mathrm{\mu g} \mathrm{~g}^{-1}$ fresh mass was 1.96 (95\% CI 1.19, 3.24) times higher than quantities in September roots, and 2.07 (95\% CI 1.23, 3.49) time higher than amounts in November roots ( $\mathrm{t}_{50.6}=2.7, P=0.009$, and $\mathrm{t}_{41.3}=2.81, P=0.008$, respectively). There was no difference ( $\mathrm{t}_{50.9}=0.19, P=0.850$ ) in xylem propanol quantities between roots sampled in September and November ( 0.30 and $0.28 \mu g^{-1}$ fresh mass, respectively).

## A1.2. Sites 1, 2 and 3, 4 DBH and radial growth

At sites 1 and 2 the mean DBH was 45.4 ( $\pm 1.3 \mathrm{SE}$ ) cm and 42.8 ( $\pm 1.4 \mathrm{SE}$ ) cm with mean 5year radial tree growth of $1.4( \pm 0.1 \mathrm{SE}) \mathrm{cm}$ and 2.1 ( $\pm 0.1 \mathrm{SE}) \mathrm{cm}$, respectively. At sites 3 and 4 the mean DBH was $40.8( \pm 1.4 \mathrm{SE}) \mathrm{cm}$ and 32.9 ( $\pm 1.9 \mathrm{SE}$ ) cm with mean 5-year radial tree
growth of 1.9 ( $\pm 0.1 \mathrm{SE}) \mathrm{cm}$ and 2.2 ( $\pm 0.1 \mathrm{SE}$ ) cm, respectively. Diseased and healthy trees were pooled across all sites for statistical analysis, with no differences in their DBH or growth (Appendix Table A2, A3).


Fig. A1. Sites 1, 2 mean (95\% CI) water contents in Douglas-fir root phloem and xylem for the main effects of tree condition $\mathrm{A}, \mathrm{B}$ ) and month sampled C, D). Bars with the same letters are not statistically different. For graphs showing no statistical differences, see Table A1 for model $P$ values.

Table A1 Site 1, 2 statistical analysis results for various headspace volatiles and water contents in root phloem and xylem from diseased and healthy trees

| Volatile | Effect ${ }^{\text {a }}$ | Phloem |  |  | Xylem |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | d.f. | $F$ | $P$ | d.f. | $F$ | $P$ |
| Acetaldehyde | Tc | 1, 58.9 | 17.50 | <0.001 | 1, 42.5 | 26.48 | <0.001 |
|  | M | 2, 46.7 | 73.59 | <0.001 | 2, 46.6 | 3.25 | 0.048 |
|  | Tc x M | 2, 46.7 | 2.24 | 0.118 | 2, 46.6 | 1.96 | 0.153 |
| Methanol | Tc | 1, 30.6 | 2.53 | 0.122 | 1, 1.6 | 10.39 | 0.115 |
|  | M | 2, 39.4 | 1.98 | 0.152 | 2, 45.5 | 1.10 | 0.342 |
|  | Tc x M | 2, 39.4 | 0.62 | 0.543 | 2, 45.5 | 5.32 | 0.008 |
| Propanol | Tc | 1, 35.0 | 10.65 | 0.003 | 1, 38.3 | 8.63 | 0.006 |
|  | M | 2, 47.4 | 0.69 | 0.508 | 2, 46.8 | 5.37 | 0.008 |
|  | Tc x M | 2, 47.4 | 0.01 | 0.988 | 2, 46.8 | 1.79 | 0.179 |
| Water | Tc | 1, 1.0 | 0.17 | 0.749 | 1, 1.0 | 0.14 | 0.774 |
|  | M | 2, 45.7 | 25.98 | <0.001 | 2, 38.7 | 2.61 | 0.087 |
|  | Tc x M | 2, 45.7 | 0.89 | 0.416 | 2, 38.7 | 1.49 | 0.239 |

${ }^{\mathrm{a}} \mathrm{Tc}=$ tree condition (diseased, healthy), $\mathrm{M}=$ month

Table A2 Statistical analysis results for DBH and growth of diseased and healthy trees pooled across all four sites, 1-4

|  | DBH |  |  |  | Growth |  |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| Effect $^{\mathrm{a}}$ | d.f. | $F$ | $P$ |  | d.f. | $F$ | $P$ |
| Tc | 1,116 | 0.00 | 0.983 |  | 1,115 | 0.00 | 0.963 |

Table A3 DBH and growth of diseased and healthy trees pooled across all four sites, 1-4

| Tree <br> condition | DBH cm | Growth cm |
| :--- | :---: | :---: |
| Diseased | $40.3(32.2,48.5)^{\text {a }}$ | $1.9(1.3,2.4)$ |
| Healthy | $40.4(32.0,48.8)$ | $1.9(1.3,2.4)$ |
| ${ }^{\text {( }}$ (95\% CI) |  |  |

