**Supplemental material S3** 

Time-kinetics of volatiles released during light-dark transients from aspen (*Populus tremula*) leaves of different age

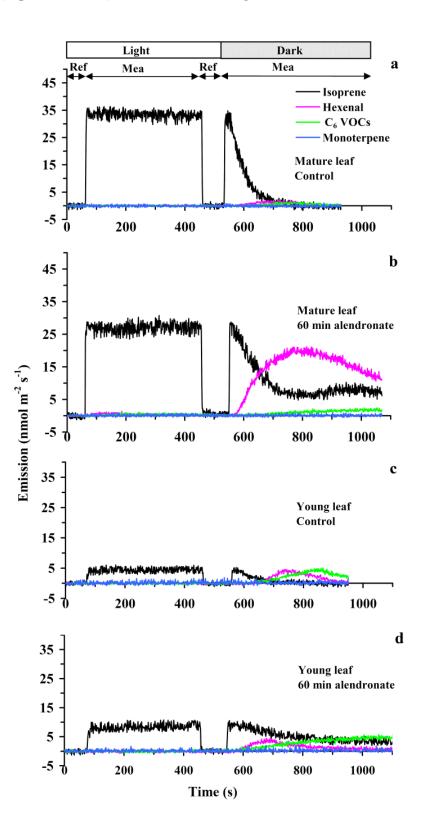


Figure S3.1 Illustration of the kinetics of volatiles released during light-dark transients from mature (a, b) and young (c, d) control (a, c) and alendronate-treated aspen (Populus tremula) leaves. The transients demonstrate periods of measurement of reference lines (Ref) and foliage (Mea). Alendronate is a specific inhibitor of prenyltransferases, in particular, geranyl diphosphate (Lange et al., 2001; Burke et al., 2004) and farnesyl diphosphate synthase (Bergstrom et al., 2000; Burke et al., 2004). The leaf was maintained at a quantum flux density of 650  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>, leaf temperature of 30 °C and ambient CO<sub>2</sub> concentration of 360 µmol mol<sup>-1</sup> until a steady-state isoprene emission rate was observed. At t = 550 s, light was switched off, leaf was simultaneously returned to the reference channel and dark release of volatiles was measured. The measurements were conducted with a high-sensitivity protontransfer reaction mass spectrometer (PTR-MS) and in addition to protonated isoprene (m/z = 69), also masses corresponding to monoterpenes (m/z = 137) and lipoxygenase pathway volatiles (LOX), main fragments of hexenals (m/z = 81) and other C6 volatiles (m/z = 83, sum of hexenols, hexanal, and hexenyl acetates) were measured following Graus et al. (2004). Dark release of these lipoxygenase volatiles has been studied in detail in grey poplar (P. x canescens) and suggested to reflect disturbances generated by light-dark transients with corresponding release of free polyunsaturated fatty acids and their oxidation by lipoxygenases (Graus et al., 2004). The kinetics and magnitude of the release of LOX volatiles from the control leaves in our study broadly corresponds to the results of Graus et al. (2004), but the LOX emissions in alendronate-inhibited leaves were larger and continued for a longer period.

## References

- Bergstrom JD, Bostedor RG, Masarachia PJ, Reszka AA, Rodan G (2000) Alendronate is a specific, nanomolar inhibitor of farnesyl diphosphate synthase. Archives of Biochemistry and Biophysics 373: 231-241
- Burke C, Klettke K, Croteau R (2004) Heteromeric geranyl diphosphate synthase from mint: construction of a functional fusion protein and inhibition by bisphosphonate substrate analogs. Archives of Biochemistry and Biophysics 422: 52-60
- Graus M, Schnitzler J-P, Hansel A, Cojocariu C, Rennenberg H, Wisthaler A, Kreuzwieser J (2004) Transient release of oxygenated volatile organic compounds during light-dark transitions in grey poplar leaves. Plant Physiology 135: 1967-1975

Lange BM, Ketchum REB, Croteau RB (2001) Isoprenoid biosynthesis. Metabolite profiling of peppermint oil gland secretory cells and application to herbicide target analysis. Plant Physiology 127: 305-314