

Time-since-death, decay classes and decay progression of deadwood in beech and silver fir forests in the central Apennines (Italy)





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Introduction

The greater part of studies on deadwood decay dynamics have been carried out in forests of North America or Northern Europe





Olympic National Park, Port Angeles, US

In Europe, especially in Mediterranean ecosystems, little is known about these processes, in part because centuries of logging have dramatically reduced the deadwood amount of original forests.



Abeti Soprani, Silver fir relict forest, Italy



Introduction

Decay state of dead wood:

visual aspects of wood, according to the decay class systems;

- qualitative evaluation of the degradation of the wood, but not an objective description of the progression rate of wood decay;

Decay Class	Structural Integrity	Texture of Rotten Portions	
1	Sound, intact, freshly fallen, bark on	Intact, no rot	
2	Sound	Mostly intact, sapwood rotting and soft, can't pull off easily	
3	Heartwood sound, supports own weight	Heartwood hard, rot beginning, large cubical rot pieces, sapwood easily pulled off or missing	
4	Heartwood rotten, does not support its own weight, but holds shape	Heartwood soft, small cubical decay pieces, metal pin easily pushed into heartwood	
5	None! Spreads out on ground, losing shape of log	Wood is soft, crumbly, heavily decomposed, powdery when dry	

Role of dead wood in carbon storage, carbon cycle and biodiversity conservation

Reliable estimates of decay rates for different tree species are needed

Objectives

 Tree-ring analyses and dendrochronological techniques have been used to measure the timesince-death of silver fir and beech stumps naturally died;



Test an objective method for tracking the rate of progression of wood decay and to understand the relationship between the time-since-death of stumps and several morphological characteristics typically described in the field;

Assess changes in chemical variables (Lignin, Cellulose, C and N content) in deadwood during the decomposition in relation to decay classes;



		Study area	Abeti Soprani	Montedimezzo
	Molise District, Italy	Latitude (degrees)	41° 51' 51''	41° 45' 16''
		Longitude (degrees)	14° 17' 54''	14° 16' 05''
		Altitude (m a.s.l.)	1260,00	1100,00
		Aspect	North	North
A. Alba	a site	Slope (degrees)	10-15 °	10 °
	F. Sylvatica site	Annual T° mean	8.4 °C	8.6°C
F. Svlva		Annual pp mean (mm)	1124,00	1022,00
		Forest type (EEA 2007)	Mediterranean and Anatolian fir forest	Apennine Corsican mountainous beech



Forests unmanaged since1960



Beech site (high forest): "*Montedimezzo*" MaB Unesco Reserve

Silver fir site: "*Abeti Soprani*", Natura 2000 site, relict stand of past glacial eve

- Cross-sections of Fagus sylvatica (54) and Abies alba (45) from stumps naturally died;
- Cores from dominant and co-dominant living trees: *Abies alba*: 16 trees

Fagus sylvatica: 15 trees



Freshly Fallen: Class 1

The assignment of each stump to a decay class in the field was based on a visual assessment, considering three classes of decay, according to the five-class system described by Hunter (1990)



Moderate Decay: Class 2



Mostly Decayed: Class 3

- Standard dendrochronological methods were used to build a tree averaged series and the two mean site chronologies (Frittz 1976)

Ring widths were measured to the nearest 0.01 mm using the LINTAB-measurement equipment, coupled to a stereomicroscope (60x magnification; Leica, Germany).



Raw ring widths of the single series of each dated trees were plotted, checked visually and then cross-dated statistically by the per cent agreement in the signs of the first-differences of the two time series (the Gleichlaufigkeit, GLK) (Kaennel and Schweingruber 1995)



The year of death of each stump was determined by identifying the calendar year in which the outermost ring of each tree was formed, matching a ring-width series from dead crosssections of unknown date of death to the dated site chronology.

To understand the relationship between time-since-death and decay classes, the average of the year of death for each class of decay in both species was compared.



Methods Lignin Extraction: from samples of all the five decay classes

Method of Niklas et al., 2000:

- Ethanol extraction;
- Water extraction;
- Acid extraction.





Freeze 1g of sample in liquid nitrogen and homogenize for 60s with mortar and pestle, Add 10ml homogenization buffer (50mM Tris-HCl, 10g/L Triton X-100, 1M NaCl pH8.3).

Wash pellet twice with 4ml of homogenization buffer, Wash pellet twice with 4ml of 80% acetone, Wash pellet twice with 4ml of pure acetone and aliquot in eppendorf.

Treat pellet with 0.4mL of thioglycolic acid + 2mL of 2M HCl for 4h at 95°C in oven.

Wash three times with distilled water, Extract lignothioglycolic acid from each pellet with 2mL of 0.5M NaOH agitating for 16h at 20°C, Centrifuge and collect supernatants.

Measure absorbance against a NaOH blank at 280nm, express lignin content as a percentage.

Cellulose Extraction: from samples of all the five decay classes

Method of Ritter G.J. and Kurth E.F., 1933, modified by Anne Kress (Laboratory of Atmospheric Chemistry (LAC), established 1 January 2000, is a laboratory of the General Energy Research Department (ENE) at the Paul Scherrer Institute, Villigen, Switzerland).

For extracting cellulose, the fibrous materials are reacted with a chemical disintegration solution in the presence of organosilicic compounds;

- **1.** Sodium Chlorite (NaClO₂), to eliminate lignin;
- 2. Sodium Hydroxide, to remove fats, resins, oils, tannins and hemicelluloses;
- 3. Hydrogen chloride;





C and N content: from samples of all the five decay classes

Total C and N of wood samples were determined with a CN auto analyzer (NA 1500, Carlo Erba Instruments).



The Carlo-Erba NA 1500 analyzer is an instrument designed for the simultaneous determination of total nitrogen and carbon in a wide range of organic and inorganic samples.



The method is based on the complete and instantaneous oxidation of the sample by "flash combustion" which converts all organic and inorganic substances into combustion products.

The resulting combustion gases pass through a reduction furnace and are swept into the chromatographic column by the carrier gas which is helium.

The gases are separated in the column and detected by the thermal conductivity detector which gives an output signal proportional to the concentration of the individual components of the mixture.

Statistical analyses

The Time Series Analysis Programme (TSAP) software package (Frank Rinn, Heidelberg, Germany) was used for statistical analysis.

GLK: measure of the year-to-year agreement between the interval trends of two chronologies based upon the sign of agreement, or the sum of the equal slope intervals in percent.

GSL: statistical significance of the GLK: ***99.9%; **99%; *95%.

TVBP: Student's t-value to investigate the significance of the best match identified. It determines the degree of correlation between curves, eliminating low-frequency variations in the time series.

The distribution of each population was tested using the Kolmogorov-Smirnov normality test (Dagnelie 1973, 1975).

Two-sample t-tests and non-parametric Mann-Whitney tests were used on independent samples in order to compare decay classes.

Results: Crossdating techniques efficiency



Tree-ring chronologies of decay classes and the mean chronology for living trees



A. alba: dated 77% of total stumps sampled

Significant correlation coefficients with reference chronologies were found for curves of single stumps and a year of death was assigned to 99 sampled stumps.

F. sylvatica: dated 69% of total stumps sampled

Results: years since death high variability in the transition rates from one class to the next



F. sylvatica and *A. alba* tend to decompose at the same rate with the exception of the third class, in which *F. sylvatica* needed more time to pass from the second to the third class;

The correlation was not strong and the age variation of stumps within decay classes was rather high;

Additional information are required to discern if the decay is the effect or the cause of the death.



Abies alba:

- lignin in living wood higher than deadwood;
- decay classes 1 ÷ 4: similar amounts;
- decay class 5: lignin significantly lower;

Fagus sylvatica:

- lignin in living wood similar to deadwood in classes 1 ÷ 3;

- decay classes from 3 to 5: lignin reduces significantly;



- decay classes 1 ÷ 3: similar amounts;

reduces significantly;

- decay classes from 3 to 5: cellulose

Abies alba:

- decay classes 1, 2: similar amounts;
- decay classes from 3 to 5: cellulose reduces significantly;



For both species, a significant increase of N in the most decayed wood occurs (classes 4 & 5)

Increased N due to fungi N import??

Fragmented material more accessible to microflora (Sollins et al., 1987); the microbial biomass increased with advancing decay, and bacterial N-fixation may have occurred within the rotting logs (Lambert et al., 1980).



-The organic C concentrations in decaying wood did not vary significantly throughout the decay process, for both species, ranging between 44% and 46% of total dry mass;

- These results are similar to those measured for coniferous species (Ganjegunte et al., 2004; Lambert et al., 1980; Sollins et al., 1987).

Conclusions (1)

- Results support previous suggestions that dendrochronological methods are useful for estimating tree mortality (Daniels et al. 1997; Hennon et al. 1990; Huggard 1999; Mast and Veblen 1994; Rouvinen 2002)

- The correlation between decay classes and the number of years since tree death was not strong and the age variation of stumps within decay classes was rather high;

- Several stumps may have died and continued decaying for many years, but the last living section of the stem may have stopped functioning rather recently. Therefore, some features of a log could erroneously indicate advanced stages of decay, and mask a recent tree death;

- Unfortunately, we were unable to identify if the stumps analyzed were the result of breaks or derived from trees that had died years before falling down. In order to understand the relationship between the year of death and the class of decay better, the cause of death of each tree should be known;

-Probably, additional information are required to discern if the decay is the effect or the cause of the death;

Conclusions (2)

- The relatively slower degradation of lignin compared to cellulose indicates that lignin can act as an important long-term source of soil organic carbon (Ganjegunte et al., 2004);

- A number of factors may have contributed to the N increase during decay processes:

1) fungi probably retained the N of the original substrate in the shrinking wood mass through immobilization;

2) the microbial biomass increased with advancing decay and bacterial N-fixation may have occurred within the rotting logs;

- Organic C concentrations in decaying wood is constant throughout the decay process, demonstrating the important role of deadwood as C-sink;

- Finally, biological processes and the role of soil fauna (earthworms and microarthropods) and soil microorganisms, especially fungi, still need to be investigated in Mediterranean mountainous forest ecosystems.



