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Disturbance of forest by trampling: Effects on mycorrhizal roots of seedlings and mature trees of *Fagus sylvatica*

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Abstract

The effects of disturbance by recreational activities (trampling) on changes in soil organic matter (SOM) and on mycorrhizal roots of seedlings and mature trees were studied in four stands of a beech (*Fagus sylvatica* L.) forest near Basel, Switzerland. At each site, comparable disturbed and undisturbed plots were selected. Disturbance reduced ground cover vegetation and leaf litter. Beech seedlings had lower biomass after disturbance. Ergosterol concentration in seedling roots, an indicator of mycorrhizal fungi, was lower in two of the four disturbed plots compared to undisturbed plots; these two disturbed sites had especially low litter levels. Based on ergosterol measurements, mycorrhizas of mature trees did not appear to be negatively affected by trampling. Total fine roots and SOM were higher in the disturbed than in the undisturbed plots at three sites. At the fourth site, fine roots and SOM in the disturbed areas were lower than in the undisturbed areas most probably due to nutrient input following picnic activities. Principal component analysis revealed a close correlation between SOM and fine roots of mature trees as well as litter and seedling biomass. Trampling due to recreational activities caused considerable damage to the vegetation layer and in particular to the beech seedlings and their mycorrhizal fine roots, whereas, roots of mature trees were apparently resilient to trampling.

Introduction

Forests are of great ecological, aesthetic, and economic importance to Switzerland (Brassel and Brändli, 1999). Visitation of forests near cities is steadily increasing and the various recreational interests such as playing with children, hiking, jogging, picnicking, dog walking, horse riding are causing trampling in areas aside paths and recreation areas. The extent of damage to the understorey increases with number of forest visitors; both the percentage of ground cover and the number of plant species decrease (Cole, 1987; Rusterholz et al., 2000). A gradient of damage to the vegetation occurs from the entry points into forests to the more distant areas (Rusterholz et al., 2000).

Forest ecosystems, including the indigenous wildlife from large animals and trees to soil microorganisms, have a limited tolerance of disturbances (Baur et al., 1999; Kuss and Morgan, 1984; Larsen, 1995). The level of disturbance that can be tolerated without exceeding capacity of the physical and biotic system to regenerate needs to be quantified in each case (Cole, 1995a,b; Marshall, 2000; Norton, 1996). Management policies can then be implemented to avoid severe, long-lasting and perhaps irreparable damage such as erosion and loss of valuable species in an area.

Forest trees of the temperate and boreal climate zone live in symbiosis with ectomycorrhizal fungi

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(Smith and Read, 1997). In this mutualistic symbiosis, photosynthetically fixed carbon from the plants is exchanged for minerals taken up from the soil by the hyphae of the ectomycorrhizal fungi. The ectomycorrhizal fungi function in the acquisition of mineral nutrients during the productive phase of ecosystems but their main role is played during the protective phase of ecosystems in the final stage of the succession when most resources are incorporated into biomass (Pankow et al., 1991). In this stage, ectomycorrhizas short-circuit nutrient cycles by direct re-acquisition of nutrients in organic form from plant litter, preventing loss of resources from the entire ecosystems. Such a short-circuited nutrient cycling was demonstrated in a model system with *Betula pendula* seedlings which acquired nitrogen and phosphate from beech-, birch-, and pine litter directly through their mycorrhizal fungi (Perez-Moreno and Rea, 2000). This model also revealed the importance of the litter layer for seedling establishment.

Little is known about trampling and ectomycorrhizas, but the effects of soil compaction on woody plants have received considerable attention (Koszlowski, 1999; Ungar and Kaspar, 1994). In general, heavy machinery, pedestrian traffic and trampling by animals lead to soil compaction, and the ensuing alterations in soil structure and hydrology negatively affect plant root growth.

With regard to mycorrhizas, studies on soil compaction have focussed mainly on arbuscular mycorrhizas in agricultural systems (Kozlowski, 1999). As recently reviewed (Augé, 2001a), the arbuscular mycorrhizal symbiosis typically affects soil structure (Miller et al., 1995; Tisdall et al., 1997), which in turn affects water retention properties in a beneficial way for plants (Augé et al., 2001b). The mycorrhizal fungus, *Glomus mosseae*, was able to absorb phosphate at high bulk density even under conditions of reduced root growth in a model system with *Trifolium pratense* (Li et al., 1997). Mycorrhizal growth response occurred at bulk densities from 1.2 to 1.6 g cm^{-3} for *Trifolium subterraneum* colonized by *Glomus* species but not at higher soil densities (Nadian et al., 1998). The symbiosis was also heavily affected by soil compaction as both the total length of the colonized root and root biomass decreased above a critical value of pressure both in *Liriodendron tulipifera* and *Liquidambar styraciflua* seedlings, and *Zea mays* and *Trifolium incarnatum* (Simmons and Pope, 1988; Entry et al., 1996).

Undisturbed forest soils typically have high porosity and low bulk densities. Soil compaction, due to site preparation and stump removal, reduced the number and morphological types of ectomycorrhizas on Douglas-fir seedlings (Amaranthus et al., 1996; Harvey et al., 1996; Page-Dumroese et al., 1998). In contrast, ectomycorrhizas of western white pine seedlings were not affected by soil compaction (Page-Dumroese et al., 1998).

Because of the central role of fine roots and ectomycorrhizas for the nutrition of trees and thus for the functioning of a forest ecosystem, a field trial in the forest of Allschwil near Basel, Switzerland, was established in areas with increasing disturbance due to recreational activities. The question was whether trampling causes physical and biological changes in soil that affect the ectomycorrhizal fine roots of beech seedlings and mature trees in this forest ecosystem.

Materials and methods

Study sites and experimental design

Four study sites (50 m × 100 m) were chosen in the forest of Allschwil situated at the southwestern edge of the city of Basel, Switzerland. Baur et al. (1999) recently described the tree community in the forest of Allschwil in detail. Beech (*Fagus sylvatica* L.) would be the dominant, naturally occurring tree species, but in many places it has been replaced by other trees, mainly *Quercus petraea, Quercus robur, Fraxinus excelsior* and *Carpinus betulus*. Considering the entire area, beech trees cover 11% of the forest of Allschwil. However, in the areas considered in the present study, beech is the dominant tree species. This forest is exposed to recreational activities. The study sites were selected to represent a rather flat area which is homogeneous with respect to management, forest tree (*F. sylvatica*), understorey and soil type (alkaline calcareous loamy sandy Flurisol). In four sites, two areas with contrasting levels of disturbance due to recreational activities were designated as a weakly disturbed ‘control’ plot and a strongly disturbed ‘trampled’ plot, each measuring 10 × 10 m. Thus, the present study is based on eight plots (four disturbed and control plots) in four sites. Hereafter, sites are referred to by their local names, i.e. Dorenbach, Bannplatz, Wasserturm and Mühlirain. Each plot was chosen such that an adult 60 to 80-year-old beech marked its centre. In each plot, samples of beech

Table 1. Species composition and cover of ground layer and shrub layer (%) in disturbed and control areas at four sites[a]

	Dorenbach		Bannplatz		Wasserturm		Mühlirain	
	Disturbed	Control	Disturbed	Control	Disturbed	Control	Disturbed	Control
Herb layer:								
Grasses and sedges:								
Carex sylvatica				1		1–5	+	1–5
Juncus svlvatica				1				
Poa annua			1–5					
Herbs and tree seedlings:								
Acer platanoides	+ −1	1–5		+	1	1–5		1–5
Ajuga reptans								+
Anemone nemorosa	+ −1	1–5	1	1–5	1–5	1–5	1–5	5–10
Asperula odoratum						1–5		
Carpinus betulus	+ −1			1–5		1–5		1–5
Circaea lutetiana	+			+			+	+
Fagus sylvatica		1–5		1–5	+	5–10		1–5
Fraxinus excelsior		+		1–5	+	1–5		1–5
Galium cruciatum		+						
Geum urbanum				+		++		++
Hedera helix	+ −1	1–5		1–5	1–5	5–10		1–5
Impatiens parviflorum				1–5				
Lamiastrum galeobdolon								1–5
Maianthemum bifolium								+
Oxalis acetosella				1–5				
Phyteuma spicatum							1	
Polygonatum multiflorum							1–5	1
Ranunculus auricomus							1–5	1
Ranunculus ficaria	1	10–20	1–5	10–20	1–5	1–5	1–5	1–5
Rubus sp.				5–10	+	+		
Rumex sp.				+				
Sorbus torminalis						1–5		
Veronica sp.								+
Viola reichenbachiana				+		1		+
Shrub layer:								
Acer platanoides		10		+	1	1–5		1–5
Carpinus betulus				1–5		1–5		1–5
Fagus sylvatica				1–5	+	5–10		1–5
Fraxinus excelsior		+		1–5	+	1–5		1–5
Crataegus laevigata					+	+		

[a] + indicates 1 individual, ++ 2 or 3 individuals.

seedlings, litter, and soil were taken in the year 2000. Soil coring was performed at 4–5 m distance from the focal beech stem.

Vegetation mapping

In each plot, the composition and abundance of plants of the herb layer were assessed in six subplots (measuring 1 m × 1 m) in April 2000 using the Domin-Krijina method (Müller-Dombois and Ellenberg, 1974). The composition and cover of the shrub layer was recorded for the entire plot. Species composition and cover of the herb and shrub layer in disturbed and control plots for the four study sites are listed in Table 1.

Sampling of seedlings and fine roots of adult trees

Seedlings of *F. sylvatica* were counted in six permanently marked subplots (1 m × 1 m) on each plot in spring and autumn. On each plot, six randomly selected seedlings were excavated in May 2000. They were washed and freeze dried for 48 h. Dry weights of the roots and shoots were determined. Afterwards, the roots were milled (Ball-mill, Retsch, MM2224, Haan, Germany) and the fine powder was stored at −20 °C.

Sampling of fine roots of adult trees and soil was carried out on three occasions: in spring (2–11 May), summer (17–22 July) and autumn (11–16 September). During each sampling period and in each plot, 10 randomly chosen soil cores of 30 mm diameter were obtained using a soil corer after removal of the litter layer (Humax, Luzern, Switzerland). The soil cores were immediately wrapped into plastic cases and processed according to Wiemken et al. (2001a). Cores were taken to a depth of 200 mm, but data are shown only for the uppermost soil layer of 100 mm depth. Fine root biomass in the lower layer of the core (100–200 mm depth) did not differ significantly between control and disturbed plots. On the average, in the lower layer fine root biomass was around 30% less than in the upper layer.

Ergosterol determination

Ergosterol, a fungus-specific sterol, was measured in roots to assess fungal biomass associated with fine roots. The measurement of ergosterol concentration in natural substrates is the most efficient method for estimating fungal biomass in temperate soils (Ruzicka et al., 2000), even more so in fine roots of trees with a high enrichment of living ectomycorrhizal fungi (Ekblad et al., 1998; Plassard et al., 2000; Wiemken et al., 2001b). Aliquots of 30 mg milled fine roots were extracted with 500 μl of 100% methanol in a sonication bath at 50 °C for 30 min. After centrifugation (10 min, 13,000 rpm), 50 μl of supernatant was analysed for ergosterol with a reversed-phase high-pressure liquid chromatography column (solid phase, Nova-Pak C 18, Waters Milford, MA, USA; liquid phase, gradient of 90–100% aqueous methanol) (Martin et al., 1990; Wiemken et al. 2001b).

Soil analysis

For the analysis of soil properties, four randomly chosen samples of 180–200 cm^3 of the topsoil layer (depth 0–100 mm, excluding the litter layer) were collected from each plot at the end of May. The four samples from each plot were pooled, mixed and sieved (mesh 4 mm). Five g of soil (dry weight) were shaken in 50 ml extraction medium for 2 h and then filtered. Extraction media were: sodium acetate (0.1 *M* for phosphate and potassium), D-lactic acid (0.02 *M* Ca-lactate in 0.02 *M* hydrochloric acid) for Mg, and hydrochloric acid and sulfuric acid (0.05 N/0.025N) for Fe, Mn and Zn (Anonymous, 1997) (Table 2). Cations were determined by atomic absorption spectrophotometry, and phosphate was determined colorimetrically (molybdate-phosphate complex) (Anonymous, 1997). The amount of litter was estimated in five randomly chosen subplots (300 mm × 300 mm) in each plot in June 2000. After oven drying at 80 °C for 48 h, the material was separated into woody parts, branches, twigs were removed and leaf litter was weighed. Soil organic matter was determined as loss on ignition of oven-dried soil at 550 °C for 24 h. Ambient soil water content (%) was determined from fresh weight/dry weight ratios of 30 g fresh soil. Soil bulk density was calculated using the dry weight/volume ratios of the top layer (100 mm depth).

Statistical analysis

The following experimental design was chosen: two treatments (trampled, control) with four repetitions (sites), ten samples and nine variables (ergosterol concentration extracted from tree fine roots and from seedling roots, fine root-, seedling-, litter-biomass, seedling number, SOM, vegetation cover). Exceptions are mentioned in figures and in the specific section. Data are presented as means ± SE. Pair-wise differences were analysed by a *t*-test. One-way analysis and repeated measure analysis of variance (ANOVA) were used to assess the effects of recreational activities (disturbed/control plots). Principal component analysis (scatter plot) was used to examine whether disturbed and control plots at the four sites differ on the basis of the nine variables. A correlation matrix PCA (correlation circle) was also calculated to reveal relations between the nine variables. Software packages JMP from SAS Institute Inc. (Cary, NC, USA) and ADE-4 (PCA; Thioulouse et al., 1997) were used to perform the analyses.

Table 2. Plant available minerals (mg 100 g^{-1} dry soil) in control and disturbed areas at the four study sites in the forest of Allschwil. Samples from the topsoil layer (0–100 mm) were taken in May 2000

Parameter	Site							
	Dorenbach		Bannplatz		Wasserturm		Mühlirain	
	Control	Disturbed	Control	Disturbed	Control	Disturbed	Control	Disturbed
pH (H_2O)	6.7	7.5	5.1	6.4	6.5	6.6	7.6	7.2
Phosphorus (P)	1.3	3.7	3.7	3.3	3.7	4.8	2.8	3.5
Potassium (K)	4.2	3.3	3.3	6.6	6.6	11.6	4.6	5.0
Magnesium (Mg)	11	7	6	7	31	22	22	20
Copper (Cu)	0.19	0.02	0.18	0.34	0.25	0.60	0.02	0.05
Iron (Fe)	10.2	0.2	25.3	55	9.4	16.5	0.2	1.5
Manganese (Mn)	9.6	3.2	9.1	19.4	22.7	25.8	4.8	18.1
Zinc (Zn)	0.6	0.2	0.5	0.8	1.2	0.8	0.1	1.0
Calcium (Ca)	155	1183	220	118	340	333	1198	1087

Results

Effects of trampling on the herb layer and the shrub layer vegetation

Disturbed plots contained fewer plant species in the herb layer than undisturbed plots in early April 2000. Furthermore, disturbed plots showed a significantly reduced plant cover (Table 3). The species and their percentage of cover are listed in the Table 1. In the control plots, 15–35% of the area was covered by shrubs. In contrast, the shrub layer was completely destroyed in the disturbed plots at three of the four sites. Thus, the ground was almost free of vegetation in disturbed plots (Figure 1).

Effects of trampling on seedlings of F. sylvatica *and its ectomycorrhizas*

The number of tree seedlings was reduced in three disturbed plots, compared to control plots, by the first sampling in spring (Table 4). A repeated survey of the same plots revealed that 100% of the seedlings had died in the disturbed plots of the Dorenbach and Bannplatz sites, whereas, the corresponding control plots showed seedling mortalities of 78 and 33%. At the other two sites the seedlings had a similar mortality in disturbed and in undisturbed plots (Wasserturm: undisturbed 40%, disturbed: 33%; Mühlirain: undisturbed 90%, disturbed: 100%). The mean biomass of a seedling was significantly lower on disturbed plots than on undisturbed plots (Tables 4 and 7). Both shoots and roots were affected in the same way, i.e. the shoot/root ratio was not altered.

Ergosterol (μg ergosterol/g DW root) was used to estimate ectomycorrhiza infection of *F. sylvatica* seedlings (Table 4). Disturbed plots had a reduced ergosterol concentration at two sites (Dorenbach: decrease 66% p=0.002, Bannplatz: decrease 38%, p=0.175). At the other two sites the ergosterol concentration did not differ significantly between disturbed and control plots. Ergosterol concentration varied significantly among study sites and was affected differently at sites as indicated by the significant interaction term between treatment and study sites (Tables 4 and 7).

Effects of trampling on fine root biomass of F. sylvatica *and its ectomycorrhizas*

When evaluated by repeated measure ANOVA, the fine root biomass was higher in disturbed plots than in undisturbed plots (Table 7). Pair-wise comparisons (Table 5) showed this effect to occur in five out of six data pairs for the Dorenbach and Bannplatz sites. In contrast, at the Wasserturm site, fine root biomass was lower in disturbed plots than in undisturbed plots in each of the three seasons. No significant difference was observed at the Mühlirain site.

Ergosterol as a marker for ectomycorrhiza formation tended to be higher in summer at all sites. Interestingly, the study sites differed significantly in ergosterol concentration (‘fungal biomass’) while there was no effect of trampling (Table 7). The Dorenbach site had especially high ergosterol levels in undisturbed plots in autumn and the Mühlirain site in disturbed plots in the same season.

Figure 1. The Bannplatz site. Top: control plot with densely growing understorey, bottom: disturbed plot with almost bare soil.

Table 3. Plant species richness and percentage cover of herb layer and shrub layer[a]

Parameter	Site							
	Dorenbach		Bannplatz		Wasserturm		Mühlirain	
	Control	Disturbed	Control	Disturbed	Control	Disturbed	Control	Disturbed
Herb layer: Number of species	5.5±0.2	2.5±0.4	9.5±0.7	2.7±0.3	9.2±0.6	4.5±1.0	10±0.6	5.8±0.6
Cover (%)	26±2.6	4.2±1.2	44±8.1	3.0±1.5	41±6.0	11±3.5	34±6.5	4.6±0.8
Shrub layer: Number of species	4	0	4	0	5	3	7	0
Cover (%)	35	0	15	0	25	5	25	0

[a]Data for the herb layer are means per m^2 ± SE, n=6, data for the shrub layer represent single measurements for the whole plots.

Table 4. Beech seedlings collected at the four study sites in spring 2000[a]

Parameter	Site							
	Dorenbach		Bannplatz		Wasserturm		Mühlirain	
	Control	Disturbed	Control	Disturbed	Control	Disturbed	Control	Disturbed
Number of seedlings	70±16	**4**±1	13±6	**2**±1	45±14	**0.3**±0.3	126±23	75±27
Seedling biomass	312±27	**212**±15	319±2	**234**±24	293±12	**227**±18	284±26	**174**±16
Root biomass	65±4	**36**±4	63±11	53±7	52±7	41±6	59±6	**32**±3
Shoot biomass	247±24	**176**±14	255±17	**181**±21	241±7	**186**±14	225±22	**142**±14
Ergosterol	145±24	**50**±4	88±18	55±14	68±10	72±17	49±6	47±11

[a]Number of seedlings per m^2, biomass of seedlings, roots, shoots (all in mg dry weight per seedling) and ergosterol concentration (μg per g DW root) as fungal marker. Data are means ±SE, n=6. Bold numbers indicate data that are significantly different from the corresponding control according to a t-test. See Table 7 for further statistical analysis.

Effects of trampling on soil characteristics

Bulk density was significantly higher in disturbed plots than in undisturbed ones (Table 7). Out of twelve pair-wise comparisons, only three showed significantly higher soil densities at disturbed sites, and one showed a significantly lower soil density at the disturbed site (Table 6). In contrast to the differences in amount of litter and soil organic matter, the differences in soil density were small, ranging from 0 to 20% (Table 6). The highest value measured was 1.54 g cm^{-3} (disturbed plot at Bannplatz site).

The litter biomass was significantly lower in the disturbed plots compared to control plots (Tables 6 and 7). In two of the four sites (Dorenbach, Bannplatz) the litter biomass in disturbed plots was more than 70% lower than the control. At the other two sites (Mühlirain, Wasserturm), it was reduced by 50 and 22%, respectively. Comparing the undisturbed plots, the lowest litter biomass was found in the Mühlirain site. Considering disturbed plots exclusively, the highest litter biomass was also recorded in the Mühlirain site. This suggests that the extent of disturbance by recreational activities is lower at the Mühlirain site than at the other three sites.

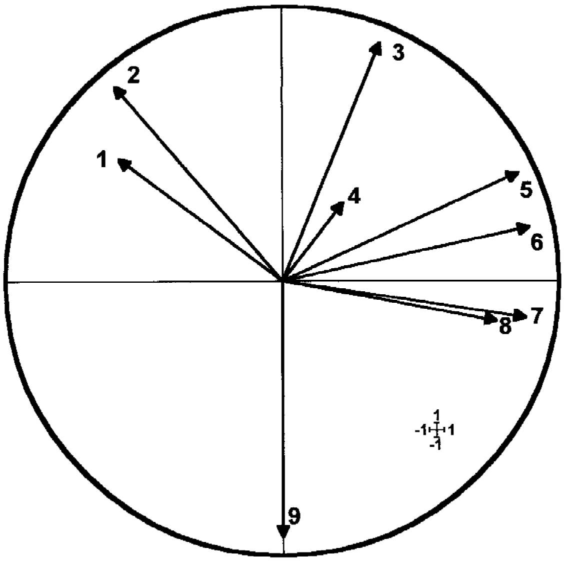

Figure 2. Correlation circle of the PCA with the nine variables. Clockwise (1) fine root biomass, (2) soil organic matter, (3) seedling number, (4) ergosterol concentration extracted from tree fine roots, (5) litter biomass, (6) percentage of vegetation cover, (7) seedling biomass, (8) ergosterol concentration of seedling roots, (9) soil density.

Table 5. Fine root biomass (DW per 100 ml soil) at four sites determined on three seasons and fungal biomass (μg ergosterol/g fine root DW)[a]

Parameter Season	Site Dorenbach		Bannplatz		Wasserturm		Mühlirain	
	Control	Disturbed	Control	Disturbed	Control	Disturbed	Control	Disturbed
Fine root biomass								
Spring	386±32	**555**±45	211±35	280±67	487±43	**223**±46	318±36	357±76
Summer	294±33	**447**±67	143±34	**447**±67	363±45	**230**±27	308±41	348±40
Autumn	324±58	**479**±60	126±24	**326**±72	432±48	**186**±44	288±41	381±49
Fungal biomass (ergosterol concentration)								
Spring	123±14	**52**±7	77±15	69±9	68±8	74±12	66±7	122±30
Summer	159±37	145±70	93±20	124±23	93±15	169±47	69±10	138±36
Autumn	273±32	**67**±12	115±29	71±10	74±9	85±15	127±25	147±22

[a] Samples were taken by soil coring and analysis of the top layer (100 mm). Data represent means ±SE (n=10). Bold numbers indicate data that are significantly different from the corresponding control according to a t-test. See Table 7 for further statistical analysis.

Soil organic matter was between 20 and 39% higher in the disturbed than in the control plots at three sites, while at the Wasserturm site, it tended to be lower in the disturbed than in the control plots (Tables 6 and 7). Soil organic matter showed little variation over the three seasons at each individual plot. Ambient soil water content did not differ between disturbed and control plots at the four sites (Table 7). Pair-wise comparisons revealed three exceptions (Table 6). Thus, in general, water relations did not appear to contribute to the differences between control and disturbed plots. Correlation matrix PCA with vectors representing the nine variables studied showed a close relationship between soil organic matter and fine root biomass, as both vectors point in the same direction and are of similar length (Figure 2). Another close relationship exists between seedling ergosterol and seedling biomass and a weaker relationship with litter biomass and percentage of vegetation cover. In contrast, soil bulk density was not related to any of the measured parameters (Fig. 2). Univariate statistics: soil bulk density/ergosterol: r^2=0.16, p=n.s; soil bulk density / SOM: r^2=0.60, p<0.024; soil bulk density / fine root biomass: r^2=0.05, p=n.s.

Comparison of the plots by principal component analysis

In order to compare the plots, PCA was performed on all parameters examined (Figure 3). The analysis separated disturbed and control plots on the PC axis 1 (inertia 42%), indicating a major effect of trampling. In addition, three control and three disturbed plots were separated on PC axis 2 (inertia 38%). The disturbed plot at the Mühlirain (M2) site and the control plot at the site Bannplatz (B1) were separated from the other three disturbed and control plots, respectively.

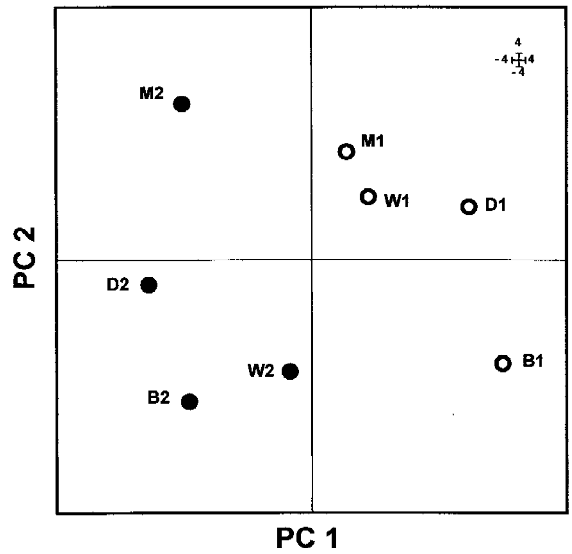

Figure 3. Scatter plot of the PCA with the four study sites and disturbed and undisturbed plots. PC axis 1 and PC axis 2 explain together 73% of the inertia. 1 = control, 2 = disturbed; B = Bannplatz, D = Dorenbach, M = Mühlirain, W = Wasserturm.

Discussion

In the urban regions of central Europe, forests are very frequently subjected to trampling by recreation activities, particularly during the warm seasons. These activities reduce the cover of the herb and shrub layer,

Table 6. Biomass of soil litter, soil organic matter (SOM), ambient soil water content and soil density of the top layer at the four investigation sites, as measured during spring, summer and autumn season[a]

Parameter	Site							
Season	Dorenbach		Bannplatz		Wasserturm		Mühlirain	
	Control	Disturbed	Control	Disturbed	Control	Disturbed	Control	Disturbed
Soil density (g cm^{-3})								
Spring	1.23±.07	1.25±.06	1.35±.03	1.38±.06	1.35±.06	1.44±.06	1.32±.07	**1.12**±.05
Summer	1.20±.03	1.28±.04	1.24±.05	**1.40**±.05	1.13±.03	**1.30**±.05	1.01±.04	1.04±.03
Autumn	1.25±.04	1.25±.04	1.33±.07	**1.54**±.06	1.24±.04	1.34±.05	1.09±.06	1.09±.05
Litter (g m^{-2})								
Spring	794±103	**225**±13	913±125	**202**±23	896±100	**451**±44	679±55	**481**±83
Spring	6.3±0.3	**8.6**±0.2	4.9±0.2	**6.3**±0.5	9.0±0.5	7.8±0.3	8.0±0.5	**12.6**±1.1
Summer	6.4±0.4	**8.4**±0.2	4.9±0.2	**6.3**±0.5	9.5±0.4	**7.4**±0.4	11±0.5	**14**±0.9
Autumn	6.5±0.3	**8.9**±0.3	5.2±0.6	5.4±0.4	9.0±0.2	7.9±0.5	10±0.5	**14**±0.7
Water content (%)								
Spring	23±1	23±1	22±1	23±1	24±1	23±1	24±1	27±2
Summer	17±1	18±1	19±1	**13**±1	22±1	19±1	23±1	23±2
Autumn	11±1	**14**±1	18±1	**10**±0.4	16±1	16±1	18±1	19±1

[a] For soil litter and SOM the difference between disturbed and control plots are given additionally in percent. Data are means ±SE, n=10. Bold numbers indicate data that are significantly different from the corresponding control according to a t-test. See Table 7 for further statistical analysis.

Table 7. Summary of p-values (based on least-square means) in the ANOVAs that examine the effects of trampling (T) and study site (S) on plant parameters (see Table 4) and fine roots and fungal biomass of *F. sylvatica* (see Table 5) and soil parameters (see Table 6)

Response variable	ANOVA type	Source of variation (p-values)		
		Trampling (T)	Study site (S)	Interaction (T×S)
Seedlings (Table 4)				
Number	two-factor	0.0001	0.0001	0.019
Biomass	two-factor	0.0001	0.15	0.74
Root biomass	two-factor	0.0001	0.19	0.30
Shoot biomass	two-factor	0.0001	0.19	0.88
Ergosterol	two-factor	0.0015	0.011	0.042
Trees (Table 5)				
Fine root biomass	repeated measure	0.034	0.0001	0.0001
Fungal biomass	repeated measure	0.085	0.024	0.0001
Soil parameters (Table 6)				
Litter	repeated measure	0.0001	0.0004	0.0001
SOM	repeated measure	0.0001	0.0001	0.0001
Bulk density	repeated measure	0.0076	0.0001	0.0071
Soil water	repeated measure	0.078	0.0001	0.0001

exposing bare ground to erosion and degradation that concerns forest practitioners and conservation biologists alike (Cole, 1987, 1995a,b; Rusterholz et al., 2000). These signs of damage to the physical and biological integrity of the ecosystem may indicate that the vitality and even the survival of the adult trees and even more so the seedling is threatened. At our study sites in the forest of Allschwil, the first signs of damage were apparent after only a few years of recreational use. Damage to understorey by trampling reduced the litter layer and the establishment of tree seedlings including their ectomycorrhizas. Thus, loss of litter is of great concern regarding seedling establishment, but ultimately also regarding the health of the mature trees which depend on ectomycorrhizas for nutrient acquisition from the litter layer.

In previous studies, the understorey recovered at formerly frequently visited sites within 4 years when the visitors were excluded by a fence, but plant species diversity was still low (Rusterholz, unpublished data). *Pinus edulis* seedlings were reduced by 73% in heavily tramped areas compared to lightly trampled areas in the Garden of the Gods, Colorado springs, Colorado (Tonnesen and Ebersole, 1997). Seedlings were less affected near woody plants due to a more even microclimate in summer as well as in winter and protection from direct injury of trampling. Demarcation of trek routes and their rotational use helped revitalise the colonisation of understory vegetation in a city forest in central Japan (Bhuju and Ohasawa, 1998). In one study, the mortality reached almost 100% for many species of the understorey vegetation in forests with only 100 to 300 trample incidents (Kuss and Hall, 1991). However, protection of the sensitive plants from trampling allowed almost immediate recovery.

After trampling, soil organic matter (5–14%) remained within the normal range for forest soils. The soil organic matter was significantly higher in the top-100-mm soil layer in the trampled plots at three of the four study sites. This could be due to a reduced turnover or to a higher input of organic matter due to trampling.

At the Wasserturm site the SOM was reduced at the trampled plot as compared to the control plot, probably because of a more rapid turnover of the organic matter. This site also had a lower fine root biomass in the trampled plots. These changes were likely due to a high input of mineral fertilizer at this site heavily used as a picnic area. The changes observed between trampled and undisturbed sites were apparently not related to soil compaction. The maximal bulk density of a soil sample monitored was 1.54 g cm^{-3}, i.e. below the value of 1.6 g cm^{-3} considered critical in soil compaction studies not only for roots but even for fungal hyphae (Nadian et al., 1998; Schachtschabel et al., 1992). Trampling did not affect the water content of the soil.

The number of beech seedlings was strongly reduced on the trampled plots as compared to the control plots at three of the four study sites, and seedlings persisting on the trampled plots had a reduced shoot biomass at all four sites, indicative of damage due to trampling. Most likely, such damage was the cause of the high mortality and lower biomass rather than soil compaction. At two of the sites (Dorenbach and Bannplatz), ectomycorrhiza formation of the seedlings, as estimated by the ergosterol content, was lower in the trampled plots than in the control plots, while at the two other sites (Wasserturm and Mühlirain) mycorrhiza formation in the seedlings was not affected. At the latter sites, trampling caused much less reduction of the leaf litter than at the former ones, indicating that a well-developed litter layer may be important for mycorrhizal colonization of the seedlings and ultimately for a well-developed community of ectomycorrhizal fungi.

Mature trees had a significantly increased fine root biomass at disturbed sites, while the fungal biomass did not change significantly. This may indicate that the trampling positively affected root but not fungal growth (Table 6). In other studies of ectomycorrhizas a concomitant increase of beech and spruce fine roots and the fungal biomass attached to the roots was found at elevated atmospheric CO_2 in calcareous soil (Wiemken et al., 2001a,b). This was also true for spruce seedlings and the ectomycorrhizal fungus *Pisolithus tinctorius* in a model system (Ineichen et al., 1995).

While the roots of adult trees appear resilient to the trampling conditions, seedlings and ectomycorrhizal fungi of mature forest trees appear to suffer on the sites where the litter layer is stongly reduced, potentially affecting the whole ecosystem in the long run.

Disturbed sites of the forest of Allschwil could be improved by simple management strategies. Any tools preventing mechanical damage to the understorey would allow re-establishment of normal forest on the disturbed plots since the soil did not appear to be heavily compacted or damaged. This became obvious by comparing soil densities, soil ambient content, and fine root biomass of disturbed and undisturbed plots for the different sites. Sustainable recreational use of the forest of Allschwil would need a management that conserves a normal litter layer as a key condition for high soil quality and thus good health of trees and shrubs. Many small animals would benefit from such a management. Visitors can be restricted to forest roads and paths by various natural obstacles: (1) logs can be placed along forest roads, (2) cut branches can be piled up along paths and prevent trespassing, (3) harvested trees can be cut at a height of 1 m instead of soil level which allows fast growth of bushes and young trees around the protecting trunk. Such stumps are barriers against human trampling and provide additional habitats for fungi, small animals, mosses and ferns. (4) Young *Picea abies* and *Abies alba* develop naturally as understorey in many forests with old beech as main tree where light is limited for deciduous bushes and trees. Instead of cutting these unwanted species, they could be left as barriers for up to an age of 15 years providing shelter for mammals as deer, badger, fox, and hare. A well-developed and esthetical path system is of course the prerequisite of all such management practises.

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