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Author(s)	Saitoh, Seikoh; Mizuta, Hitomi; Hishi, Takuo; Tsukamoto, Jiro; Kaneko, Nobuhiro.; Takeda, Hiroshi
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特集　ニホンジカの森林生態系へのインパクト—芦生研究林

論　　文

Impacts of deer overabundance on soil macro-invertebrates in a cool temperate forest in Japan: a long-term study

S. Saitoh*, H. Mizuta*, T. Hishi**, J. Tsukamoto***, N. Kaneko****, and H. Takeda*

日本列島の冷温帯林における 30 年間の土壌動物相の変化とシカ類の影響の可能性

齋藤　星耕＊・水田　瞳＊・菱　拓雄＊＊・塚本　次郎＊＊＊・金子　信博＊＊＊＊・武田　博清＊

Deer overabundance affects many taxa and ecosystem functions in both aboveground and belowground systems. We investigated changes in soil macrofauna along a ridge and at the bottom of a slope in Ashiu Experimental Forest, a cool temperate natural forest in central Japan, in which the flora, fauna, microflora, and ecosystem functions were studied before the Sika-deer *(Cervus nippon)* population increased. The abundance and biomass of soil macrofaunal detritivores (earthworms, isopods, amphipods, and diplopods) and predators (opilionids, Araneae, and chilopods) were adversely affected both along the ridge and at the slope bottom. These results are consistent with prior studies conducted with deer exclosures or natural experiments. The soil organic matter distribution, the carbon-nitrogen ratio in the litter and mineral layers, and soil hardness along the slope also changed. These results suggest that deer overabundance affects ecosystem functions along the slope. The movement of litter and soil along the slope, and the reduction of soil macrofauna associated with decomposition possibly contributed to these changes..
Key words: cool temperate forest, overabundance, Sika-deer, soil macrofauna

温帯におけるシカ類の密度の増大が、広範な植物相と動物相、生態系プロセスに影響することが示されてきた。我々はニホンジカ (Cervus nippon) の増加以前の土壌動物相の知見が蓄積されている京都大学芦生演習林（中部日本）において、斜面の尾根部と谷部において大型土壌動物相を調査して過去のデータと比較した。過去と現在の間の変化は尾根と谷とで類似しており、腐植食者のミミズ類、等脚類、端脚類、倍脚類と、捕食者であるイシムカデ類、ジムカデ類、クモ類、ザトウムシ類が個体数や現存量に負の影響を受けていた。尾根部で地表徘徊性甲虫の密度が増加していた。我々の大型土壌動物に関する結果は排除柵や自然の実験を用いた、有蹄類が土壌動物に及ぼす影響に関する先行研究の結果によく一致するものであった。同じ期間に斜面における有機物土壌の分布と、表層リターおよび土壌における C/N 比、土壌硬度が顕著に変化しており、シカが生態系機能に影響を与えていることが示唆された。斜面上の有機物の移動や分解者としての土壌動物相の減少がこの変化に寄与した可能性がある。
キーワード：土壌動物，ニホンジカ，食害

Introduction

In the past several decades, changes in land use, lack of predators, declining hunting pressure, and low mortality in moderate winters have resulted in overabundance of cervid populations in boreal and temperate forests in developed countries (reviewed by Côté et al., 2004; Stewart, 2001; Latham et al., 2005). A rapid increase of Sika-deer *(Cervus nippon)* populations is a problem in Japan (Hino et al., 2003). Deer overabundance degrades horizontal and vertical diversity in plant communities, and thereby has negative effects on bird, small mammal, and invertebrate diversity (Latham et al., 2005).

Aboveground herbivores can indirectly affect belowground systems by affecting plant physiology or the community, or both (Wardle, 2002). As "keystone herbivores," ungulates often change the flora in a forest (reviewed by Côté et al., 2004). Selective grazing by overabundant ungulates clearly alters the biomass and

* Graduate School of Agriculture, Kyoto University
** Faculty of Agriculture, Kyushu University
*** Faculty of Agriculture, Kochi University
**** Graduate School of Environment and Information Sciences, Yokohama National University
* 京都大学大学院農学研究科地域環境科学専攻
** 九州大学農学部
*** 高知大学農学部
**** 横浜国立大学大学院環境情報研究院

species composition of plant communities (e.g., Molvar et al., 1993; Pastor et al., 1993). Plant species that are unpalatable to ungulates often have high concentrations of secondary metabolites or indigestible polymers such as lignin in their tissues (Côté et al., 2004). Therefore, a plant community exposed to high ungulate grazing pressure provides the belowground system with less labile carbon and nutrient sources. On the other hand, ungulates provide dung and urine as labile carbon and nitrogen sources for the soil surface, and contribute to soil processes (Frank and Groffman, 1988; Pastor et al., 1993; Haynes and Williams, 1999; Williams et al., 2000). Furthermore, intensive grazing reduces understory foliage and inhibits regeneration of canopy trees (Takatsuki and Gorai, 1994), resulting in changes to the forest floor microclimate, i.e., wind speed, the diurnal amplitude in temperature, and dry-wet frequency (Stewart, 2001). These changes affect the soil microflora and fauna that are responsible for decomposition. Thus, ungulates affect ecosystem functions via multiple mechanisms (Bardgett and Wardle, 2003).

The effects of ungulates on soil biota vary depending on the size class of soil organisms. While the microbe-protozoa-nematode system responds differently based on nutrient conditions or trophic structure (reviewed by Wardle, 2002), the abundance and biomass of larger soil animals, including meso- and macrofauna, are largely negatively affected in heavily grazed forests (Suominen, 1999; Wardle et al., 2001; Allombert et al., 2005; Ito,

unpublished data) and grasslands (e.g., Clapperton et al., 2002; Petersen et al., 2004), except for some ground-dwelling insects (Suominen et al., 1999a, 1999b, 2003; Allombert et al., 2005).

Prior studies were conducted using exclosure experiments, in which large herbivorous mammals were excluded from plots, or natural experiments, in which faunal communities under different grazing intensities or with different histories were compared. To determine whether predictions based on these prior studies can be applied to a cool temperate forest in Japan, we investigated the soil macrofauna and compared it with that of 30 years ago on a slope in Ashiu Experimental Forest, a cool temperate natural forest in central Japan, in which the flora, soil fauna (Tsukamoto, 1977, 1985, 1986a, 1986b, 1996; Takeda, 1981; Kaneko and Takeda, 1984; Kaneko, 1985, 1988, 1989), and ecosystem functions (Katagiri and Tsutsumi, 1973, 1974, 1975, 1976, 1978; Sakai and Tsutsumi, 1986, 1987) were studied before the *C. nippon* population increased and the flora was severely damaged.

We focused on the abundance and biomass of soil macro-invertebrates to examine the impact of deer overabundance on soil fauna and soil ecosystem in a cool temperate forest with a long-term study.

Materials and methods

Study site and sampling dates
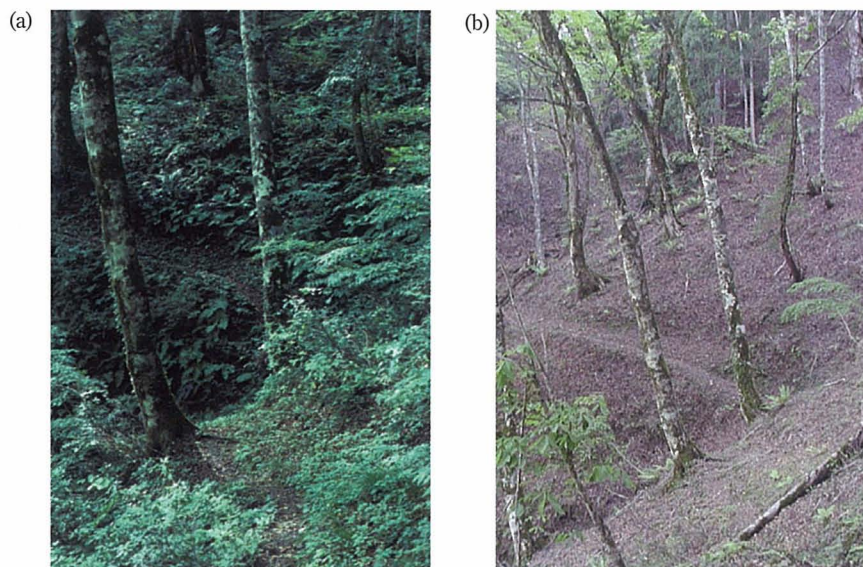
This study was conducted in Ashiu Experimental

Figure 1 Deer impacts on understory foliage in Ashiu Experimental Forest. Photographs were taken by Dr. Etsuko Katori at the same location in Iwatani Valley in (a) 1999 and (b) 2006.

Forest (35° 3′ N, 135° 8′ E), Field Science Education and Research Center, Kyoto University, located in central Honshu Island, Japan. The estimated density of *C. nippon* in the forest from 2001 to present was up to 10 individuals km⁻² (Takayanagi et al., unpublished data), equivalent or higher than the density measured at sites in earlier studies (e.g., Suominen et al., 1999a). Although no information about the abundance of deer in the 1970s was available, their density was not problematic until the late 1990s. Understory foliage in the forest has declined drastically, both in coverage and diversity (Kato and Okuyama, 2004). Figure 1 shows understory foliage at the same location in the forest in 1999 and 2006.

Almost all forests in Japan develop on slopes (Iwatsubo, 1996). The flora, distribution of organic matter, decomposition pattern, and, thus, carbon and nutrient dynamics differ along the slope (reviewed by Katagiri, 1996). Soil faunas also vary along the slope (Tsukamoto 1977, 1985, 1996; Takeda, 1981; Kaneko, 1985). Therefore, surveys both along ridges and at the bottom of slopes are needed for a detailed picture of the soil fauna in Japan. We selected a slope on the north side of a ridge in Nodahata Valley and investigated the soil macrofauna. The slope was 70 m in length and ranged from 695 to 735 m above sea level (ASL). The study site was covered with snow from December to April. Table 1 gives the cumulative precipitation and mean temperature for each month and year, measured at the Ashiu Experimental Forest office, 10 km south of the study site and 355 m ASL. On the studied slope, moder and mull soils developed along the ridge and bottom, respectively (Tsukamoto, 1977; Takeda

Table 1 Climate data for Ashiu Experimental Forest during the study period.

(a) Mean temperature (°C)

	Jun	Sep	annual
1976	19.9	18.5	11.2
1977	20.1	19.9	11.3
2007	20.2	21.1	12.2

(b) Cumulative precipitation (mm)

	Jun	Sep	annual
1976	213.0	367.0	2294.0
1977	106.0	213.0	1591.0
2007	280.5	120.5	1946.0

(a) Mean temperature and (b) cumulative precipitation by month and year measured at the Ashiu Experimental Forest office, located about 10 km south of the study site and 355 m above sea level.

and Kaneko, 1988). Both the ridge and the bottom of the slope support broad-leaved deciduous forest. *Quercus mongolica crispula* and *Lyonia ovalifolia* were dominant on the ridge and *Cornus controversa*, *Pterocarya rhoifolia*, and *Fagus crenata* were dominant at the bottom. On the forest floor, dwarf bamboo (*Sasa kurilensis*) was dominant until the late 1990s but disappeared no later than 2007 due to deer browsing.

Soil macrofauna

The soil macrofauna was investigated before deer overabundance, from 1974 to 1978 (Tsukamoto, 1977, 1985, 1986a, 1986b, 1996), and after deer overabundance, in June and September 2007. Soil invertebrates were sampled monthly from May or June to November with quadrats both along the ridge and at the bottom of studied slope in the investigation conducted in 1970s. For detailed methods of prior studies, see Tsukamoto (1977, 1985, 1986a, 1986b, 1996). The methods used in 2007 are detailed below.

In 2007, the hand-sorting method was used for soil macrofauna. Samples were collected in early June and early September 2007. We established plots with six 20-m² subplots on the ridge and slope bottom. We sampled one point randomly in each subplot with a 25 × 25 cm quadrat. Three layers, the litter layer (L layer) and two 10-cm thick layers of soil beneath the L layer, were sampled. All macro-invertebrates found in hand sorting were preserved in 70% ethanol. The animals were sorted into the following taxa or groups: large Oligochaeta (earthworms), Amphipoda, Isopoda, Diplopoda, Diptera larvae, Coleoptera larvae, Coleoptera imagoes, Chilopoda (three orders), Araneae, Opiliones, Turbellaria, and Hirudinea. Alcohol wet weight for each individual was used as the biomass indicator and was determined to 10^{-5} g with an electric balance.

The litter of the L layer was oven dried and weighed. The thickness of the FH layer (in the A_0 layer) for each pit was determined as the mean value of four replicates measured on internal aspects in the square pit after we sampled the soil. Soil hardness was measured at depths of 5, 10, and 15 cm with Yamanaka's soil hardness tester. Two soil samples taken from each pit with 100-cm³ cores at depths of 5 and 15 cm, representing the two soil layers sampled, were brought to the laboratory. We oven-dried the soil samples and sieved them with a 2-mm mesh to

remove roots and rocks. We determined total carbon and nitrogen contents of the soil and leaf litter with an NC analyzer (NC-22A, Sumigraph).

Statistical analyses

We analyzed the data obtained in 2007 and corresponding data from Tsukamoto (1977, 1985, 1986a, 1986b, 1996). The following data were used for comparison of the soil fauna before and after deer overabundance: the abundance and biomass of soil macrofauna (large Oligochaeta, Amphipoda, Isopoda, Diplopoda, Diptera larvae, Coleoptera larvae, Coleoptera imago, Chilopoda (three orders), Araneae, Opiliones, Turbellaria, and Hirudinea) in June and September 1976, 1977, and 2007.

Here, macrofauna contained in the soil organic layer (A_0 layer, containing the L and FH layers) and mineral layer (to 10 cm below the organic layer) were compared at the ridge site. At the bottom site, the macrofauna of the L layer and from below 10 cm were compared among sample dates. Note that macrofauna at the ridge site in 2007 may be overestimated by up to 15.6% for abundance and 6.4% for biomass due to a difference in sampling methods for macrofauna at the ridge site between 1976/1977 and 2007; the L layer, FH layers (ca. 3.6 cm), and below 10-cm thick mineral layer were separately sampled in 1976 and 1977, whereas the L layer and below two 10-cm thick layers, with the upper 10-cm layer containing both FH and mineral layers, were sampled in 2007 (Fig. 2). The methods used at the bottom site were the same for all sample dates. Thus, the macrofauna at the ridge site was investigated deeper in 2007 than in 1976 and 1977. In the analysis, we compared the macrofauna collected in 1976 and 1977 with all the
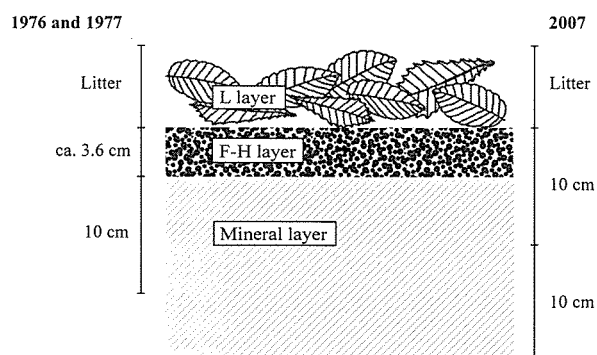

Figure 2 Sampling methods at the ridge site in 1970s and 2007. Soil strata, from which soil fauna were analyzed in present paper, are expressed with solid line on each side.

macrofauna sampled in 2007 at the ridge site; thus, animals from an extra 6.4 cm of soil were included in the 2007 data. Although the degree of overestimation caused by this was not negligible, it did not affect our conclusions, as mentioned in the discussion. Also note that examination to depths of 40 cm or deeper in soil is needed for a proper understanding of earthworms in natural forests, because many earthworms are found in deeper soil (Watanabe, 1968, 1973; Tsukamoto, 1985).

We examined whether the time elapsed from the 1970s to 2007, in which deer abundance increased, explains variation in the abundance of each taxon. We conducted a two-way analysis of variance (ANOVA) in which elapsed years and season (June or September) were the main effects. The ridge and bottom sites were analyzed separately. For taxa in which the effect of the season-time interaction was significant, the difference among samples in each season was examined by one-way ANOVA and a pair-wise test with a Bonferroni correction.

We used principal components analysis (PCA) based on faunal biomass to demonstrate changes in the macrofaunal community. Mean values of taxa or groups among replications were used in this analysis. The data were transformed by log (alcohol wet weight (mg) + 1). In accordance with the recommendation of Hasegawa (2006), detrended correspondence analysis (DCA) was run first. No gradient length in the DCA was larger than 4. Thus, we used PCA.

The following soil properties before and after deer overabundance were compared. Note that available soil property data before deer overabundance were not obtained on the same dates.

· The litter mass in the L layer in June 1976 and June 2007
· The thickness of the FH layer in June and September 1975 and June and September 2007
· The carbon and nitrogen content and C/N ratio in August 1978 and June and September 2007
· The soil hardness in August 1983 and June and September 2007

A Welch test was used to test for differences in the amount of litter, FH layer thickness, and carbon and nitrogen content. When comparing data sampled more than three times at one place and depth, a one-way ANOVA and pair-wise test with a Bonferroni correction were used. To examine changes in FH layer thickness at

the bottom site, where the FH layer was very thin in the 1970s but no thickness data for that time are available, a one sample t-test was used to determine whether the mean values in 2007 were different from zero. The correlation between soil hardness and depth in the mineral layer was tested. After that, the difference between sample dates was examined with analysis of covariance (ANCOVA), in which the data for soil hardness in June and September 2007 were pooled and compared with data from 1983.

All statistical analyses were conducted in R 2.6.1 (R Development Core Team, 2007).

Results

1. Soil properties

The amount of litter in the L layer, the thickness of the FH layer, the C/N ratio in the litter and mineral layers, and soil hardness are given in Table 2. The amount of litter accumulated in the L layer at the bottom site in 2007 was twice the level from the 1970s ($P < 0.001$); this difference was not significant at the ridge site ($P = 0.260$; Table 2.1). The thickness of the FH layer did not change significantly at the ridge site (one-way ANOVA, $P = 0.18$)

but was substantially thicker at the bottom site in 2007, where the FH layer was very thin in 1975 (Table 2.2). The C/N ratio of leaf litter in the L layer decreased at both the ridge and bottom sites, and that in the mineral layer also decreased (Table 2.3). The low litter C/N was caused by the considerably higher nitrogen concentration in litter, which was ca. 2.5–2.6% in June 2007 and 2.9–3.0% in September 2007.

At all sample dates and sites, soil hardness was positively correlated with soil depth (Pearson's product-moment coefficient test, $P < 0.001$). The ANCOVA detected no change in the slope of the regression function, where the dependent and independent variables were soil hardness and depth, respectively, at neither the ridge site ($P = 0.466$) nor the bottom site ($P = 0.814$). The adjusted mean soil hardness did not change at the ridge site ($P = 0.624$) but increased at the bottom site ($P < 0.001$). Soil hardness at a depth of 10 cm, extrapolated from the regressions, is listed in Table 2.4.

2. Soil macrofauna

Soil macrofauna abundance ($P = 0.0454$) and biomass ($P = 0.00786$) changed with time at the ridge site, and also at the bottom site (abundance, $P < 0.001$; biomass, $P <$

Table 2.1 Accumulated litter mass (g m^{-2}) in the L layer.

| | Ridge | | Bottom | |
	Mean ± SE	n	Mean ± SE	n
June 1976	366 ± 8.2	5	456 ± 58	5
June 2007	604 ± 187[ns]	6	1040 ± 141[***]	6

Values are means ± 1 SE. Significant differences between June 1976 and June 2007 were examined. * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; ns, not significant.

Table 2.2 Thickness of the FH layer (cm).

| | Ridge | | Bottom | |
	Mean ± SE	n	Mean ± SE	n
June 1975	3.4 ± 0.57[a]	5	<<1	-
Sept. 1975	3.1 ± 0.85[a]	5	<<1	-
June 2007	4.6 ± 0.64[a]	6	1.2 ± 0.26[*]	6
Sept. 2007	2.7 ± 0.92[a]	6	3.0 ± 0.78[**]	6

Values are means ± 1 SE. Different letters indicate significant differences among sample dates at the ridge site. A one sample t-test was used to determine if mean values in 2007 differed from zero for the bottom site. * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$;

Table 2.3 The C/N ratio in leaf litter and the soil layer by sample date, site, and depth.

| | | Ridge | | Bottom | |
		Mean ± SE	n	Mean ± SE	n
Leaf litter	Aug. 1978	30.3 ± 1.1[a]	4	24.1 ± 0.70[a]	4
	June 2007	22.2 ± 0.61[b]	6	21.1 ± 0.59[ab]	6
	Sept. 2007	18.9 ± 0.58[c]	6	19.3 ± 1.43[b]	6
0-10 cm	Aug. 1978	21.2 ± 0.80[a]	4	13.5 ± 0.70[a]	4
	June 2007	13.3 ± 0.57[b]	6	10.6 ± 0.25[b]	6
	Sept. 2007	13.5 ± 0.33[b]	6	10.6 ± 0.57[b]	6
10-20 cm	Aug. 1978	18.1 ± 1.0[a]	4	13.0 ± 0.90[a]	4
	June 2007	12.6 ± 0.83[b]	6	9.44 ± 0.41[b]	6
	Sept. 2007	12.9 ± 0.50[b]	6	9.26 ± 0.58[b]	5

Values are means ± 1 SE. Different letters indicate significant differences among sample dates.

Table 2.4 Soil hardness (kg cm^{-3}) at 10 cm depth.

	Ridge	Bottom
1983	0.237	0.077
2007	0.218[ns]	0.217[***]

Values were extrapolated by linear regression, where the dependent and independent variables were soil hardness and depth, respectively. ANCOVA was used to determine if adjusted mean values differed between sample dates. * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; ns, not significant.

0.001). The mean total abundance (± SE) for the pooled data from the two sample months decreased from 854.4 ± 228.5 (n = 10) in 1976 and 968.0 ± 103.9 (n = 12) in 1977 to 560.0 ± 135.4 (n = 12) individuals m^{-2} in 2007 at the ridge site, and from 1720.0 ± 252.0 (n = 12) in 1976 and 1720 ± 238.6 (n = 12) in 1977 to 512.0 ± 97.6 (n = 12) individuals m^{-2} in 2007 at the bottom site. The mean total biomass (± SE) for data pooled between the two sample months decreased from 10.7 ± 4.55 (n = 10) in 1976 and 12.4 ± 2.55 (n = 12) in 1977 to 2.22 ± 0.529 (n = 12) g m^{-2} in 2007 at the ridge site, and from 13.9 ± 2.48 (n = 12) in 1976 and 10.4 ± 1.54 (n = 12) in 1977 to 4.59 ± 2.28 (n = 12) g m^{-2} in 2007 at the bottom site.

Figure 3 shows the abundance and biomass of detritivorous macrofauna in 1976, 1977, and 2007. Earthworms decreased in abundance (P = 0.0230) and biomass (P = 0.0458) at the ridge site, and decreased in abundance (P < 0.001) but not biomass (P = 0.187) at the bottom site. Amphipoda decreased in abundance (P < 0.001) and biomass (P = 0.00152) at the bottom site. Isopoda decreased in both abundance and biomass at

both sites (P < 0.001). An effect of the season-time interaction on isopod abundance was detected at both the ridge (P = 0.0215) and bottom (P = 0.0241) sites. The abundance of Diplopoda decreased at the ridge (P = 0.0365) and bottom (P = 0.0241) sites, but biomass did not decrease at either the ridge (P = 0.301) or bottom (P = 0.237) sites. The abundance of Coleoptera imagoes increased (P = 0.0118) but their biomass did not increase (P = 0.967) at the ridge site.

Figure 4 shows the abundance and biomass of predaceous macrofauna in 1976, 1977, and 2007. Opilionid abundance marginally decreased at the ridge (P = 0.0507) and bottom (P = 0.0890) sites. Their biomass marginally decreased at the ridge site (P = 0.0516) but did not decrease at the bottom site (P = 0.191). In fact, no Opiliones individuals were collected in 2007. Araneae decreased in abundance and biomass at both the ridge (abundance, P < 0.001; biomass, P = 0.0221) and bottom (abundance, P = 0.00906; biomass, P = 0.0130) sites. In Chilopoda, Lithobiomorpha decreased in abundance and biomass at both the ridge and bottom sites (P < 0.001).

(a) Macrofaunal detritivore abundance in the ridge site   (b) Macrofaunal detritivore abundance in the bottom site

(c) Macrofaunal detritivore biomass in the ridge site   (d) Macrofaunal detritivore biomass of in the bottom site
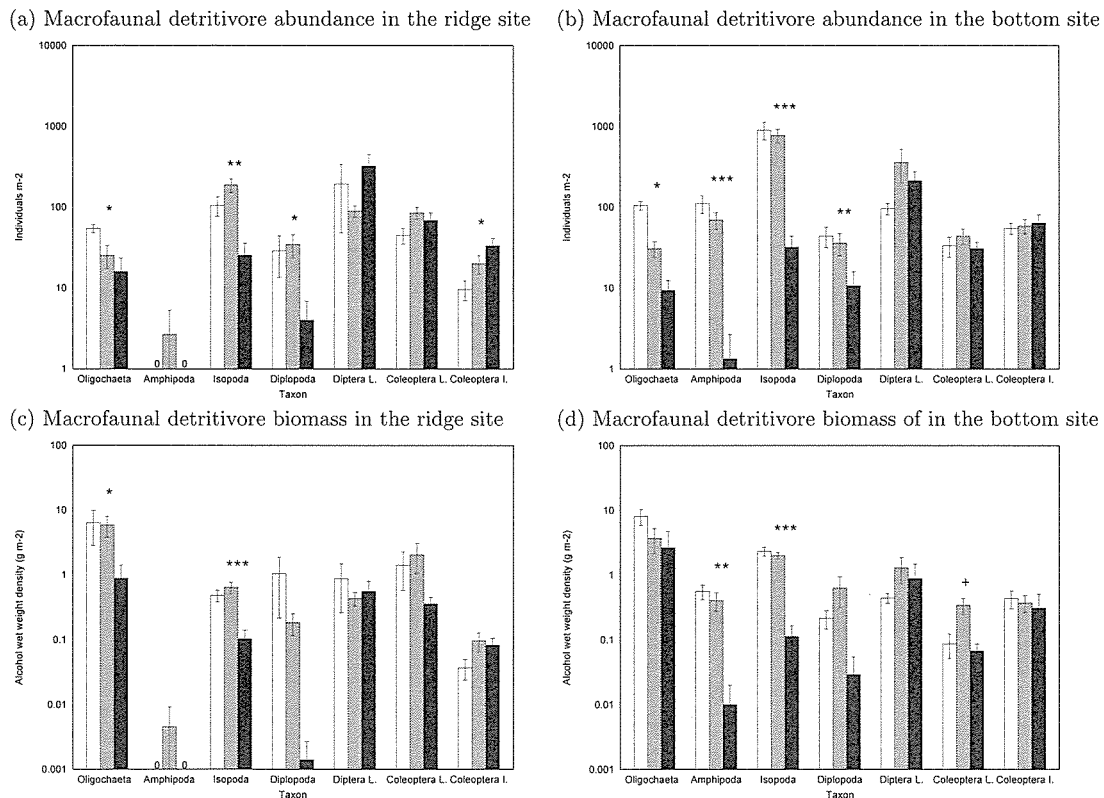

Figure 3 Abundance and biomass of macrofaunal detritivores at the ridge and bottom sites. Open, gray, and filled columns indicate abundance/ biomass in 1976, 1977, and 2007, respectively. Columns are based on pooled data from June and September (n = 12, except for the ridge site in 1976, n = 10). Bars indicate 1 SE. Symbols for taxa: I and L represent imago and larva, respectively. Note that the category "Coleoptera Imago" contains predators, herbivores, and detritivores. The following symbols indicate a statistically significant effect of time on macrofaunal abundance or biomass in the two-way ANOVA where sample month and elapsed years are the main effects. + P < 0.10; * P < 0.05; ** P < 0.01; *** P < 0.001.

(a) Macrofaunal predator abundance in the ridge site   (b) Macrofaunal predator abundance in the bottom site

(c) Macrofaunal predator biomass in the ridge site   (d) Macrofaunal predator biomass of in the bottom site
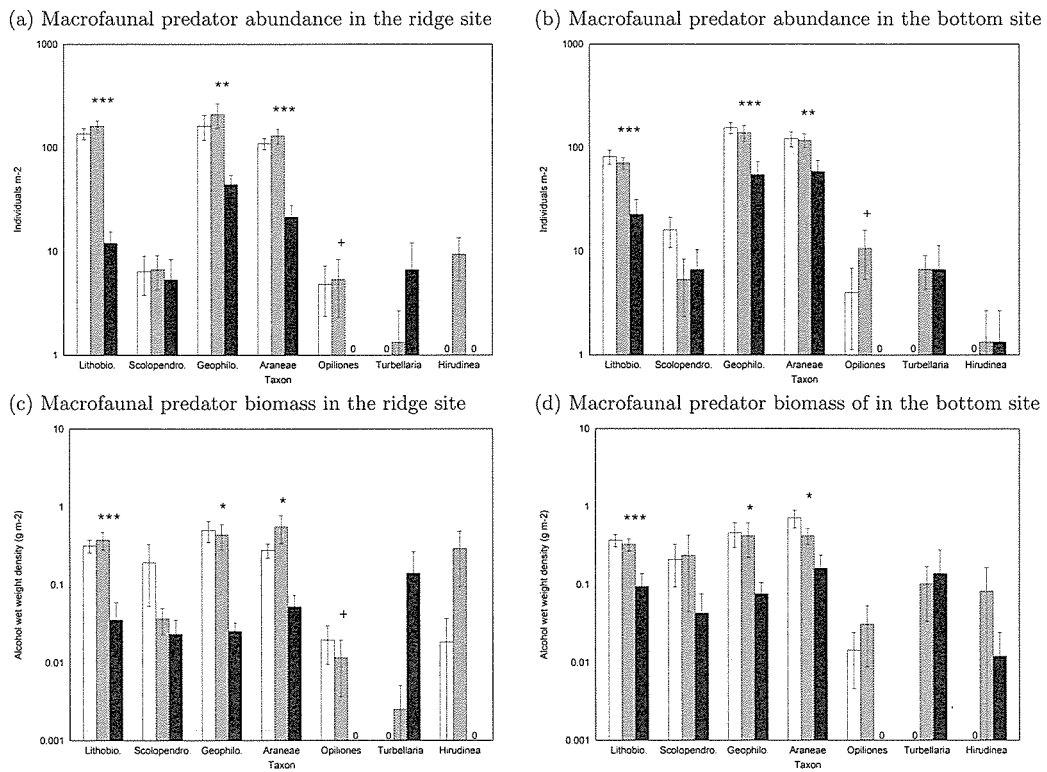

Figure 4 Abundance and biomass of macrofaunal predators at the ridge and bottom sites. Open, gray, and filled columns indicate abundance/biomass in 1976, 1977, and 2007, respectively. Columns are based on pooled data from the sampling months, June and September (n = 12, except for the ridge site in 1976, n = 10). Bars indicate 1 SE. Lithobio., Scolopendro., and Geophilo. are abbreviations for Lithobiomorpha, Scolopendromorpha, and Geophilomorpha, respectively. The following symbols indicate a statistically significant effect of time on macrofaunal abundance or biomass in the two-way ANOVA where sample month and elapsed years are the main effects. + $P < 0.10$; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

(a) Isopoda abundance in the ridge site   (b) Isopoda abundance in the bottom site

(c) Geophilomorpha abundance in the ridge site   (d) Geophilomorpha abundance in the bottom site
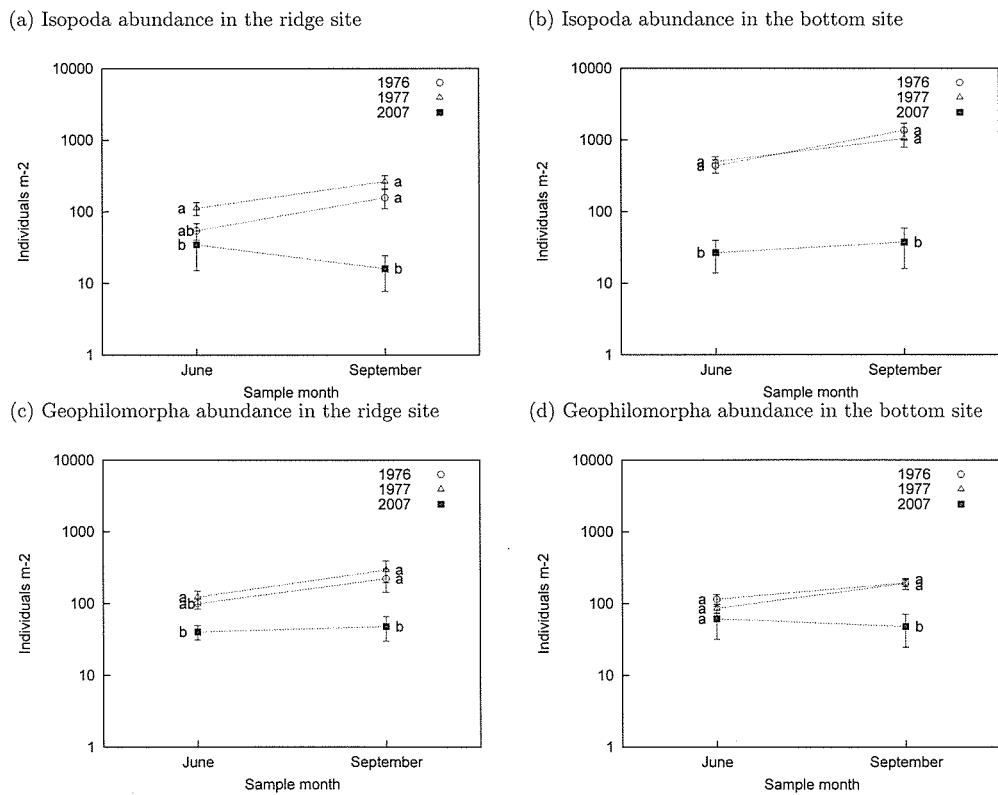

Figure 5 Seasonal change in the abundance of Isopoda and Geophilomorpha at the ridge and bottom sites. Open circles, open triangles, and filled rectangles represent abundance in 1976, 1977, and 2006, respectively. Bars indicate 1 SE (n = 6, except for the ridge site in 1976, n = 5). Different letters in the same month indicate a significant difference in a pair-wise test.

Geophilomorpha abundance and biomass decreased at both the ridge (P = 0.0040 and 0.00129, respectively) and bottom (P < 0.001 and P = 0.0486, respectively) sites. An effect of the season-time interaction on Geophilomorpha abundance was detected at the ridge site (P = 0.0215).

Figure 5 shows the abundance of Isopoda and Geophilomorpha in June and September, when the season-time interaction was significant, except for Geophilomorpha at the ridge site (P = 0.131). The difference in abundance was larger in September than in June.

Figure 6 shows the results of PCA of macrofaunal communities based on biomass. The first and second axes explained 38.7% and 22.6% of the variance among communities. Communities were divided into three groups, ridge communities in the 1970s, bottom communities in the 1970s, and all communities in 2007.

Most arrows for taxa or faunal groups are orientated in the opposite direction of arrows for 2007 communities, reflecting their reduced biomass. Coleoptera larvae and Hirudinea at the ridge site and Amphipoda at the bottom site characterized the difference between ridge and bottom communities in the 1970s.

Discussion

Impacts of deer browsing on the macrofaunal community

The total biomass of soil macrofauna decreased within 30 years at our study site. Our results show that the abundance or biomass, or both, of earthworms, Isopoda, Diplopoda, Opiliones, Araneae, and two orders of Chilopoda (Geophilomorpha and Lithobiomorpha) decreased at both the ridge and bottom sites. Amphipoda, which was abundant at the bottom site in the 1970s,
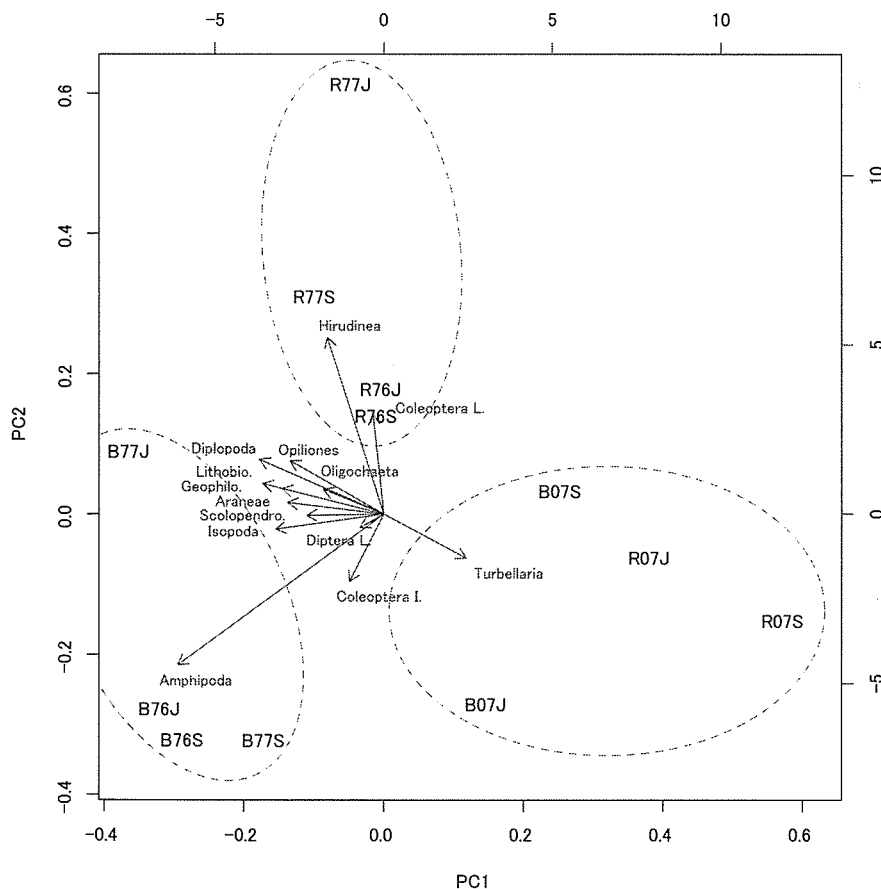

Figure 6   Ordination of the macrofaunal community based on biomass by PCA. Symbols for macrofaunal communities are as follows. The first letter (R or B) indicates if the communities were sampled at the ridge or bottom site, respectively. The numbers in the middle (76, 77, or 07) indicate if the communities were sampled in 1976, 1977, or 2007, respectively. The final letter (J or S) indicates if the communities were sampled in June or September, respectively. For instance, R76J indicates a community sampled at the ridge site in June 1976. Arrows represent faunal taxa or groups. Abbreviations are as follows. Lithobio., Lithobiomorpha; Geophilo., Geophilomorpha; Scolopendro., Scolopendromorpha; I., Imago; L., Larva..

Table 3 Responses of soil macrofaunal biomass and/or abundance to ungulate browsing in temperate and boreal forests.

Literature	Allombert et al. (2005)	Suominen (1999), Suominen et al. (1999a, 2003)[1]	R. How, unpublished data cited in Stewart (2001)	Wardle et al. (2001)	Present study
Flora type	Boreal forests	Boreal forests	Lowland deciduous woodland	Wide range of forest types	Cool temperate decidious forest (ridge and bottom on a slope)
Ungulate herbivore	Sitka black-tailed deer (Odocoileus hemionus sitkensis)	Moose (Alces alces), Reindeer (Rangifer tarandus), Roe deer (Capreolus capreolus)	Fallow deer (Dama dama)	Feral goats (Capra hircus), red deer (Cervus elaphus scoticus), fallow deer (Dama dama), white tailed deer (Odocoileus virginianus), Dama wallaby (Macropus eugenii)	Sika-deer (Cervus nippon)
Study type	Natural experiment	Exclosure experiment	Exclosure experiment	Exclosure experiment	Long-term study
Sampling methods	Pit fall trap	Pit fall trap	Pit fall trap	Extractor	Hand-sorting
Positive	Detritivores/herbivores: Curculionidae	Predators: Araneae (3 families)[2] Formicidae, Carabidae Detritivores/herbivores: Homoptera, Leiodidae, Curculionidae (in pine forests)	Predators: Carabidae (Pterostichus madidus, Nebria brevicollis)		Coleoptera (only in ridge site)
Negative	Detritivores: Gastropoda,	Predators: Araneae (2 families)[2], Opiliones, Staphylinidae Hymenoptera (excluding Formicidae) Detritivores: Gastropoda, Curculionidae (in decidious forests)	Predators: Carabidae (Abax parallelopipedus, Carabus violaceus)	Predators: Araneae, Pseudoscorpionidea, Opiliones, Chilopoda, Staphylinidae Detritivores: Gastropoda, Isopoda, Amphipoda, Diplopoda, Diptera larvae	Predators: Araneae, Opiliones, Chilopoda (Lithobiomorpha, Geophilomorpha) Detritivores: Earthworm, Isopoda, Amphipoda, Diplopoda

[1] Taxa that changed in abundance in Suominen (1999) or Suominen et al. (1999a, 2003) are listed.
[2] Total Araneae abundance was negatively affected in a pine dominated forest (Suominen et al., 1999a).

drastically declined. Ground-dwelling Coleoptera imago was the only group that increased in abundance at the ridge site.

As mentioned in the methods, the data for fauna at the ridge site in 2007 were likely overestimated due to differences in sampling procedures between the 1970s and 2007. Thus, our results should be interpreted carefully. However, our results indicate that most of the taxa investigated had declined or did not change in abundance or biomass. The abundance and biomass of ground-dwelling Coleoptera imagoes, the only group that increased in abundance at the ridge site, are unlikely to have been estimated imprecisely, because they are surface dwellers. Therefore, the possible overestimation of macrofauna at the ridge site in 2007 does not affect the discussion herein.

Table 3 compares the results of previous studies of the impacts of ungulates on soil macrofauna in forests with our results. Our findings are consistent with prior studies, in that ungulate browsing largely negatively influenced soil macrofauna, over several trophic levels (Suominen, 1999; Wardle et al., 2001), and positively influenced some groups, such as ground-dwelling Coleoptera (Suominen et al., 1999a, 2003; Allombert et al., 2005).

With deer overabundance, loss of plants that provide food and shelter for soil fauna and the resulting changes in wind speed, temperature, and moisture in the forest floor environment negatively affect animals that inhabit the forest floor (Stewart, 2001). Some of these mechanisms may apply to Ashiu Experimental Forest. Deer overabundance resulted in a drastic decline in understory coverage and flora richness in Ashiu Experimental Forest (Kato and Okuyama, 2004). We found no dwarf bamboo and the understory foliage was scarce at our study site in 2007, whereas dwarf bamboo was dominant in the 1970s. A depleted understory may have changed the forest floor microclimate and adversely affected the soil fauna. In fact, large detritivorous arthropods, such as Isopoda, Amphipoda, and Diplopoda, were drastically reduced at our study site. These arthropods are found primarily in the organic layer, feed on dead plant material, and are susceptible to desiccation (reviewed by Coleman et al., 2004).

At our study site, however, the amount of litter and the thickness of the organic layer, which provide food and habitat for such detritivores, did not decrease, and actually increased at the bottom site. Thus, it is unlikely that food depletion or desiccation occurred. This result implies that mechanisms other than those mentioned by Stewart (2001) are responsible for the faunal reduction. Tsukamoto (1977) found that two dominant species of Isopoda depend on different microhabitats within the organic layer. Although we did not evaluate the structure of microhabitats in detail, a decline in detritivorous macrofauna despite an increase in litter may indicate a change in the quality of microhabitats. Further, the difference in the abundance of Isopoda was more pronounced in September than in June (Fig. 5a, b). At the study site, isopods reproduce from June to September, and increase in abundance during that period (Tsukamoto, 1977). Thus, an impact on their recruitment processes is suggested.

The abundance and/or biomass of some macrofaunal predators (Opiliones, Araneae, and Chilopoda) also dropped. A cascade effect via detritivores may have contributed to this change (Wardle et al., 2001). The increase in Geophilomorpha, an order of Chilopoda, and Isopoda during summer was not as notable in 2007 as in the 1970s (Fig. 5c, d). Although their life histories remain to be studied, a similar mechanism may be depressing both the predatory and detritivorous soil macrofauna.

Suominen et al. (1999a, 2003) found that the abundance of some taxa increases depending on forest conditions. When deer browsing modifies the forest floor environment and enhances foliage suitable for some faunal groups, those groups can increase in abundance. In this study, only Coleoptera increased in abundance at the ridge site. At our study site, the canopy trees did not change and vegetation unpalatable to deer did not increase in the understory plant community. Future changes in plant composition may result in increases in other taxa in addition to the ground-dwelling Coleoptera.

Changes in the macrofauna over the past 30 years were similar at the ridge and bottom sites despite differences in soils. Furthermore, PCA revealed that macrofaunal communities could be classified into three groups: ridge communities in the 1970s, bottom communities in the 1970s, and all communities in 2007. These findings suggest that deer browsing depleted and homogenized the macrofaunal communities at the ridge and bottom sites. This result indicates that deer overabundance diminished beta diversity along the slope.

Impact on ecosystem processes

The amount of litter in the L layer increased at the bottom site of the study slope. Furthermore, an FH layer, which is usually found on the ridge part of a slope and not at the bottom, was newly developed at the bottom site. The amount of organic material on slopes should be a function of litter fall, decomposition rate, and in- and outflow along the slope. Complicated mechanisms may be responsible for changes in the distribution of organic material on the forest floor as follows.

First, the movement of litter and soil along the slope may be one mechanism involved in the shift in organic material distribution. Furusawa et al. (2003) demonstrated that deer browsing enhances the movement of litter and soil. On a slope, degradation of the understory loosens the soil surface and exposes it to wind. As a result, the movement of litter from the ridge and middle part of the slope to the bottom could be considerable. In fact, we observed an easily breakable soil surface, but the extent seemed to vary with microtopography.

Second, the decline of large detritivores (earthworms, Amphipoda, Isopoda, and Diplopoda) that have important roles in soil processes as ecosystem engineers or litter transformers (Lavelle, 1997; Kaneko and Itoh, 2004) may contribute to the increased amount of organic material at the bottom site. Tsukamoto (1986a, 1986b, 1996) evaluated the contribution of soil macrofauna to litter disappearance at this site, and concluded that Isopoda and earthworms were responsible for more than 30% of litter disappearance in the late 1970s. Therefore, the disappearance of litter should have been slower in 2007 due to the depleted detritivorous macrofauna.

Third, annual litter fall and decomposition rate, which basically determine the quantity of organic material that accumulates on the forest floor, may have changed. Although these parameters were not measured in the present study, the soil property results imply changes in them. The litter and soil C/N ratios were significantly lower in 2007 than in 1978 at both the ridge and bottom sites. The low litter C/N ratio in 2007 was due to an increased litter nitrogen concentration, which increased from 1.5–1.9% in 1978 to 2.5–3.0% in 2007. Studies conducted before deer overabundance showed that the nitrogen concentration of litter in the L layer at this site ranged from 1% in newly fallen leaves to at most 2% in the decomposition process (Katagiri and Tsutsumi, 1973;

Tsukamoto, 1985; Osono and Takeda, 2001). A nitrogen-rich litter may promote decomposition in early stages and retard decomposition in later stages because of inhibition of lignin decomposition by nitrogen (reviewed by Berg and McClaugherty, 2003). This might have contributed to humus accumulation, resulting in the formation of an FH layer at the bottom site. On the other hand, the low soil C/N ratio in 2007 should have promoted nitrification (Murakami et al., 1990). Therefore, NPP and, in turn, litter fall might have been enhanced (Wardle, 2002).

Excessive mineralization of nitrogen caused by ungulate overabundance could result in nutrient losses from the system (reviewed by Wardle, 2002) and thereby cause eutrophication of streams in the forest (Fujimaki et al., unpublished data).

Soil hardness increased at the bottom site, but did not change significantly at the ridge site. Physical traits of soil such as soil hardness are functions of water content, organic content, bulk density, porosity, and the extent of aggregation as a result of biological activity (Katagiri, 1996). Hence, the increased soil hardness indicates that such factors might have changed.

Our results suggest substantial changes in the material dynamics of the forest between the 1970s and 2007. However, the exact nature of these changes is not yet clear. Future studies comparing with prior data from this forest (Katagiri and Tsutsumi, 1974, 1975, 1976, 1978; Sakai and Tsutsumi, 1986, 1987) should reveal ungulate effects on ecosystem functions in cool temperate forests.

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References

1) Allombert, S., Stockton, S. and Martin, J. L. (2005) A natural experiment on the impact of overabundant deer on forest invertebrates. Conserv. Biol., 19, 1917–1929.

2) Bardgett, R. D. and Wardle, D. A. (2003) Herbivore mediated linkages between aboveground and belowground communities. Ecology, 84, 2258–2268.

3) Berg, B. and McClaugherty, C. (Eds.) (2003) Plant Litter. Decomposition, Humus Formation, Carbon Sequestration, 286 pp. Springer Verlag, Berlin, Heidelberg, Berlin, New York.

4) Clapperton, M. J., Kanashiro, D. A. and Behan-Pelletier, V. M. (2002) Changes in abundance and diversity of microarthropods associated with Fescue Prairie grazing regimes, Pedobiologia, 46, 496–511.

5) Coleman, D. C., Crossley Jr., D. A. and Hendrix P. F. (2004) Fundamentals of Soil Ecology (second ed.), 386 pp. Academic Press, San Diego.

6) Côté, S. D., Rooney, T. P., Tremblay, J. P., Dussault, C. and Waller, D. M. (2004) Ecological impacts of deer overabundance. Annu. Rev. Ecol. Evol. Syst., 35, 113–147.

7) Frank, D. A., and Groffman, P. M. (1988) Denitrification in a semi-arid grazing ecosystem. Oecologia, 117, 564–569.

8) Furusawa, H., Miyanishi, H., Kaneko, S. and Hino, T. (2003) Movement of soil and litter on the floor of a temperate mixed forest with an impoverished understory grazed by deer (*Cervus nippon centralis* Temminck). J. Jpn. For. Soc., 85, 318–325 (Japanese with English summary).

9) Hasegawa, M. (2006) Application of ordination in community ecology of soil animals. Edaphologia, 80, 35–64 (Japanese with English summary).

10) Haynes, R. J. and Williams, P. H., (1999) Influence of stock camping behaviour on the soil microbiological and biochemical properties of grazed pastoral soils. Biol. Fertil. Soils, 28, 253–258.

11) Hino, T., Furusawa, H., Ito, H., Ueda, A., Takahata, Y. and Ito, M. (2003) Forest ecosystem management based on an interaction network in Ohdaigahara. Jpn. J. Conserv. Ecol., 8, 145–158 (Japanese with English summary).

12) Iwatsubo, G. (ed.) (1996) Forest Ecology. 306 pp. Buneido Publishing, Tokyo (Japanese).

13) Kaneko, N. (1985) A comparison of oribatid mite communities in two different soil types in a cool temperate forest in Japan. Pedobiologia, 28, 255–264.

14) Kaneko, N. (1988) Feeding habits and cheliceral size of oribatid mites in cool temperate forest soils in Japan, Rev. Ecol. Biol. Sol, 25, 353–363.

15) Kaneko, N. (1989) Life histories of four oribatid mite species in a mull type soil in a cool temperate forest in Japan. Pedobiologia, 33, 117–126.

16) Kaneko, N. and Ito, M. (2004) Biodiversity and ecosystem function of soil animals. Jpn. J. Ecol., 54, 201–207 (Japanese).

17) Kaneko, N. and Takeda, H. (1984) A preliminary study on oribatid mite communities in the cool temperate forest soils developed on a slope. Bull. Kyoto Univ. For., 56, 1–10.

18) Katagiri, S. (1996) Productivity and nutrient cycling under different site conditions in forest ecosystem in Forest Ecology. Iwatsubo, G. (ed.), 306 pp. Buneido Publishing, Tokyo, 224–258 (Japanese).

19) Katagiri, S. and Tsutsumi, T. (1973) The relationship between site condition and circulation of nutrients in forest ecosystem (I): Litter fall and nutrient contents. J. Jpn. For. Soc., 55, 83–90 (Japanese with English summary).

20) Katagiri, S. and Tsutsumi, T. (1974) The relationship between site condition and circulation of nutrients in forest ecosystem (II): Moisture index as means of evaluation of site condition. J. Jpn. For. Soc., 56, 434–440 (Japanese with English summary).

21) Katagiri, S. and Tsutsumi, T. (1975) The relationship between site condition and circulation of nutrients in forest ecosystem (III): Aboveground biomass and nutrient contents of stands. J. Jpn. For. Soc., 57, 412–419 (Japanese with English summary).

22) Katagiri, S. and Tsutsumi, T. (1976) The relationship between site condition and circulation of nutrients in forest ecosystem (IV): The amount of mineral nutrient returned to forest floor. J. Jpn. For. Soc., 58, 79–85 (Japanese with English summary).

23) Katagiri, S. and Tsutsumi, T. (1978) The relationship between site condition and circulation of nutrients in forest ecosystem (V): The difference in nutrient circulation between stands located on upper part of slope and lower part of slope. J. Jpn. For. Soc., 60, 195–202 (Japanese with English summary).

24) Kato, M. and Okuyama, Y. (2004) Changes in the biodiversity of a deciduous forest ecosystem caused by an increase in the Sika deer population at Ashiu, Japan. Contrib. Biol. Lab. Kyoto Univ., 29, 433–444.

25) Latham, R. E., Beyea, J., Benner, M., Dunn, C. A., Fajvan, M. A., Freed, R. R., Grund, M., Horsley, S. B., Rhoads, A. F. and Shissler, B. P. (2005). Managing white-tailed deer in forest habitat from an ecosystem perspective: Pennsylvania case study. Audubon Pennsylvania and the Pennsylvania Habitat Alliance. Harrisburg, Pennsylvania.

26) Lavelle, P. (1997) Faunal activities and soil processes: adaptive strategies that determine ecosystem function. Adv. Ecol. Res., 27, 93–132.

27) Molvar, E. M., Bowyer, R. T. and Van Ballenberghe, V. (1993) Moose herbivory, browse quality, and nutrient cycling in an Alaskan treeline community. Ecology, 94, 472–479.

28) Murakami, M., Takeda, H. and Iwatsubo, G. (1990) Seasonal changes in nitrogen mineralization rates in the soils of *Chamaecyparis obtusa* and *Cryptomeria japonica* plantations. Bull. Kyoto Univ. For., 62, 44–54 (Japanese with English summary).

29) Osono, T. and Takeda, H. (2001) Organic chemical and nutrient dynamics in decomposing beech leaf litter in relation to fungal ingrowth and succession during 3-year decomposition processes in a cool temperate deciduous forest in Japan. Ecol. Res. 16, 649–670.

30) Pastor, J., Dewey, B., Naiman, R. J., McInnes, P. F. and Cohen, Y. (1993) Moose browsing and soil fertility in the boreal forest of Isle Royale Park. Ecology, 74, 467–480.

31) Petersen, H., Jucevica, E. and Gjelstrup, P. (2004) Long-term

changes in collembolan communities in grazed and non-grazed abandoned arable fields in Denmark. Pedobiologia, 48, 559–573.

32) Sakai, M. and Tsutsumi, T. (1986) Carbon cycles of two soil types in a cool temperate forest in Japan (I): Litter fall. J. Jpn. For. Soc., 68, 1–9. (Japanese with English summary).

33) Sakai, M. and Tsutsumi, T. (1987) Carbon cycles of two soil types in a cool temperate forest in Japan (II): Seasonal variations of soil respiration rates under the effects of soil environmental effects. J. Jpn. For. Soc., 69, 41–48 (Japanese with English summary).

34) Stewart, A. J. A. (2001) The impact of deer on lowland woodland invertebrates: a review of the evidence and priorities for future research. Forestry, 74, 259–270.

35) Suominen, O. (1999) Impact of cervid browsing and grazing on the terrestrial gastropod fauna in the boreal forests of Fennoscandia. Ecography, 22, 651–658.

36) Suominen, O., Danell, K. and Bergström, R. (1999a) Moose, trees, and ground-living invertebrates: indirect interactions in Swedish pine forests. Oikos, 84, 215–226.

37) Suominen, O., Danell, K. and Bryant, J.P. (1999b) Indirect effects of mammalian browsers on vegetation and ground-dwelling insects in an Alaskan floodplain. Ecoscience, 6, 505–510.

38) Suominen, O., Niemela, J., Martikainen, P., Niemela, P. and Kojola, I. (2003) Impact of reindeer grazing on ground-dwelling Carabidae and Curculionidae assemblages in Lapland. Ecography, 26, 503–513.

39) Takatsuki, S. and Gorai, T. (1994) Effects of Sika deer on the regeneration of a *Fagus crenata* forest on Kinkazan Island, northern Japan. Ecol. Res., 9, 115–120.

40) Takeda, H. (1981) A preliminary study on collembolan communities in a deciduous forest slope. Bull. Kyoto Univ. For., 53, 1–7.

41) Takeda, H. and Kaneko, N. (1988) Patterns of humus accumulation in forests. I. Mull and moder type humus in a broad-leaved forest. Bull. Kyoto Univ. For., 60, 33–45 (Japanese with English summary).

42) Tsukamoto, J. (1977) Soil macro-animals on a slope in a deciduous broad-leaved forest. I. Two species of terrestrial Isopoda: *Ligidium japonicum* and L. paulum. Jpn. J. Ecol., 26, 201–206.

43) Tsukamoto, J. (1985) Soil macro-animals on a slope in a deciduous broad-leaved forest. II. Earthworms of Lumbricidae and Megascolecidae. Jpn. J. Ecol., 35, 37–48.

44) Tsukamoto, J. (1986a) Soil macro-animals on a slope in a deciduous broad-leaved forest. III. Indirect evaluation of the effect of earthworms on the differential development of the A_0-layer observed between the ridge and bottom parts. Jpn. J. Ecol., 35, 601–607.

45) Tsukamoto, J. (1986b) Evaluation of contribution of earthworms to disappearance of leaf litter in forests of Japan—In comparison to those of Europe. Shinrin Ritchi (Jpn. J. For. Environ.), 28, 1–10 (Japanese with English summary).

46) Tsukamoto, J. (1996) Soil macro-invertebrates and litter disappearance in a Japanese mixed deciduous forest and comparison with European deciduous forests and tropical rainforests. Ecol. Res., 11, 35–50.

47) Wardle, D. A. (2002) Communities and ecosystems: linking the aboveground and belowground components, 392 pp. Princeton University Press, Princeton.

48) Wardle, D. A., Barker, G. M., Yeates, G. W., Bonner K. I. and Ghani, A. (2001) Introduced browsing mammals in New Zealand natural forests: aboveground and belowground consequences. Ecol. Monogr., 71, 587–614.

49) Watanabe, H. (1968) On the vertical distribution of soil macro animals in three different forests. J. Jpn. For. Soc., 50, 204–210 (Japanese with English summary).

50) Watanabe, H. (1973). Effect of stand change on soil macro animals. J. Jpn. For. Soc., 55, 291–295.

51) Williams, B. L., Grayston, S. J. and Reid E. J. (2000) Influence of synthetic sheep urine on the microbial biomass, activity and community structure in two pastures in the Scottish uplands. Plant Soil, 225, 175–185.