Impact of soil fertility and insolation on diversity of herbaceous woodland species colonizing afforestations in Muizen forest (Belgium)

Luc De Keersmaekera,*, Leen Martensb, Kris Verheyenc,
Martin Hermyc, An De Schrijverd, Noël Lustd

"Institute for Forestry and Game Management, Gaverstraat 4, B-9500 Geraardsbergen, Belgium
bInstitute of Nature Conservation, Kliniekstraat 25, B-1000 Brussels, Belgium
cLaboratory for Forest, Nature and Landscape Research, Department <!f Land Management, KU Leuven,
Vita/ Decosterstraat /02, B-3000 Leuven, Belgium
dlaboratory of Forestry, Ghent University, Geraardsbergsesteenweg 267, B-9090 Melle, Belgium

Abstract

In a chronosequence of afforestations adjacent to ancient forest, relations between herbaceous forest vegetation and possible explanatory variables, such as soil characteristics, land-use history variables and relative insolation on the forest fl.oor, were studied using linear models on species groups and a direct gradient analysis (CCA). An explorative correlation analysis indicated that the moisture, carbon, total nitrogen and phosphorus contents of the soil, were correlated with forest age. Although soit pH was not correlated with forest age, its variability was greater in ancient forest and 19th century afforestations than in recently afforested parcels. The linear models indicated that soil pH positively infl.uenced the diversity and cover of both slow-colonizing and fast-colonizing woodland species. P enrichment, in combination with a high relative insolation, stimulated *Urtica dioica* in recent afforestations. Deep shadow on the forest floor suppressed light-demanding species, such as *U. dioica,* and indirectly favored woodland species. The multivariate analysis confirmed that floristic diversity was best accounted for by the variables plant-available P, pH (KCI), nitrate (negatively correlated with insolation) and forest age. The discussion focuses on the origin of the observed variability in soil characteristics, natura! or anthropogenic, and the impact on vegetation succession. lt is concluded that most woodland species can colonize recently afforested farmland parcels in Muizen forest, as soit pH is not a limiting factor. However, P eutrophication can have an indirect negative effect, increasing competitive exclusion by *U. dioica*. Plantation of an understorey can suppress *U. dioica* and stimulate colonization of shade-tolerant woodland species in afforested farmland.

Keywords: Ancient forest plant species; Competition; Secondary succession; Soil phosphorus; Soil nitrogen; Understorey

1. Introduction

A large number of European forest plants are associated with ancient forests (Hermy et al., 1999), which indicates that forest vegetation is strongly determined by land-use history. Several studies have

*Corresponding author. Tel.: +32-54-436154;
fax: +32-54-416160.
E-mail address: luc.dekeersmaeker@lin.vlaanderen.be
(L. De Keersmaeker).

focused on the poor dispersal capacities of ancient-forest species and colonization patterns in secondary woodland (Matlack, 1994; Brunet and von Oheimb, 1998; Bossuyt et al., 1999b). The habitat quality of recent forests can also limit the colonization capacities of woodland species. The relative importance of recruitment versus dispersal limitation can vary (Verheyen and Hermy, 2001). Dzwonko and Gawronski (1994) found that the vegetation composition of 70-year-old afforestations adjacent to ancient forest was determined more by soil conditions, insolation and the influence of dominant tree species than by dispersal mode.

Following the afforestation of farmland, many soil characteristics gradually change and move towards the levels of ancient-forest soils (Goovaerts et al., 1990; Muys et al., 1992; Catt, 1994; Bossuyt et al., l999a; Verheyen et al., 1999). However, even after a long period of time, persistent differences between ancient and secondary woodland soils can be observed. Farmer arable land use generally results in increased nutrient levels, especially of P (Koemer et al., 1997; Wilson et al., 1997; Honnay et al., 1999). P is immobile and the total P content is stable (Binkley, 1986), although its availability decreases after the afforestation of farmland (Magid, 1993).

High P levels might hamper the establishment of woodland species in recent forests by increasing competitive exclusion. *Urtica dioica* benefits from a high P availability (Pigott, 1971) and therefore the vegetation in recent forests on former arable land is often dominated by this species. Hermy et al. (1993) found a negative relationship between the number of woodland species and the cover of *U. dioica*. Honnay et al. (l999) mentioned a significant negative correlation between P content and the number of ancient-forest species. Both studies faced the problem that P content and the cover of *U. dioica* were correlated with secondary forest age, and therefore it is not clear which factor (forest age or P availability) caused the low number of woodland species in recent forests.

Using field experiments in coppice woods, Pigott and Taylor (1964) demonstrated that insolation could limit the growth of *U. dioica* on fertilized soils. Therefore, a comparison of vegetation succession in afforestations with high soil P levels but varying light conditions, might provide an indication as to the impact of light-demanding, competitive species such

as *U. dioica* on the colonization rate of woodland species in afforested farmland.

We used a field study to investigate the following hypotheses: (1) soil nutrients, in particular the P content, are affected by the land-use history; (2) a high soil P availability favors competitive species (*U. dioica*); (3) woodland species benefit from shadow that reduces *U. dioica* and other light-demanding competitive species.

2. Material and methods

2.1. Study area

The Muizen forest is a 34 ha forest reserve, in a flat region at approximately 10 m above sea level, situated 15 km east of Antwerp in northern Belgium (Fig. 1). The center of the forest is somewhat lower than the periphery, but the difference in altitude does not exceed 2 m. The topsoil is of quatemary niveo-eolian origin and varies from silty sand (periphery) to sandy silt (center). In the central depression, an impermeable sandy clay layer of tertiary marine origin, at 0.5-1 m depth, impairs water percolation and creates a Gleysol (FAO et al., 1998) in the central depression (Fig. 2). The local presence of fossils in the sandy clay layer results in these Gleysols having a highly variable soil acidity, with pH (KCl) values that range between ±3 and ±7. The variability within the area delineated as Gleysol on the soil map (Fig. 2), is illustrated by Verheyen et al. (2001), who performed a continuous soil classification based on soil pH, substratum type, moisture regime and thickness of the A horizon.
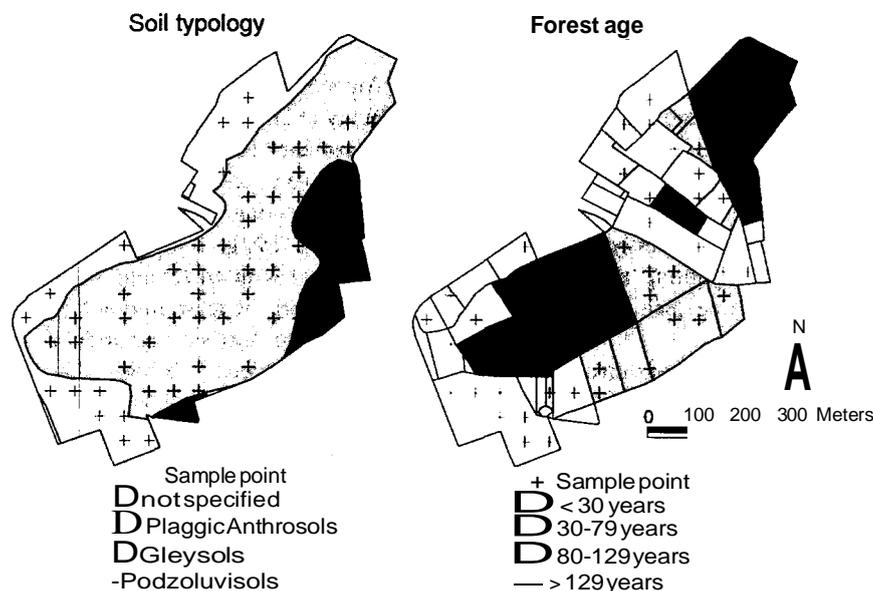

Fig. 1. Location of the study area in Belgium.

Soil typology

Sample point
D not specified
D Plaggic Anthrosols
D Gleysols
-Podzoluvisols

Forest age

N

A

0   100   200   300   Meters

+ Sample point
D < 30 years
D 30-79 years
D 80-129 years
— > 129 years

Fig. 2. Soil map and forest age map of the Muizen forest with the position of the 57 grid-based sample points.

The Gleysols are surrounded by Plaggic Anthrosols (FAO et al., 1998) in the west and by Podzoluvisols (FAO et al., 1998) in the east (Fig. 2). According to the Belgian soil map the soils are moderately wet, as depth to mottling varies from 30 to 90 cm. A frequently flooded area in the north of the forest was not included in this study.

The Muizen forest consists of a mosaic of small parcels (surface area between 0.5 and 1.5 ha) with a specific land-use history. The number of years since afforestation, further referred to as forest age, is illustrated in Fig. 2. Approximately 3.5 ha in the NE of the forest, on Gleysols and Podzoluvisols, have always been forest since 1775 when the earliest maps were drawn. This area is therefore referred to as ancient forest. Most of the Gleysols in the SW were forest in 1775 but converted into arable land between 1775 and 1834. These parcels were abandoned and reforested between 1854 and 1892. The parcels that surround the historical core of the forest, were all afforested in the 20th century. The last afforestations, largely on Plaggic Anthrosols, date from 1991.

Most forest stands were planted homogeneously with poplar cultivars on 8 m x 8 m or 1 0 m x 10 m spacing. The earliest management notes indicate that poplar cultivars had already been planted before 1928. Some clear-cuts of poplar plantations were reforested

with *Fraxinus excelsior, Tilia cordata* and *Quercus robur.* In some stands, shrubs and tree species (in particular *Acer pseudoplatanus, Alnus glutinosa* and *Corylus avellana)* were planted in between the poplars, while in others shrubs and trees established spontaneously. This resulted in varying levels of shadow on the forest floor of recent afforestations.

A wide variety of forest vegetation is found in the Muizen forest: stands where *U. dioica* or *Rubus fruticosus* are dominant neighbor stands with an abundant vemal aspect. Vegetation on acid soils belong to the *Violo-Quercetum roboris* (Oberdorfer, 1957) and vegetation on calcareous soils belong to the *Primulo-Fraxinetum excelsioris* (Henny, 1985).

2.2. Data collection

Vegetation was sampled in 57 plots of 100 m^2, set out systematically on the nodes of a 50 m x 50 m grid that covered the forest (Fig. 2). Only plots with a homogeneous vegetation were sampled. The herbaceous layer, including pteridophytes, was inventoried in April and May 1996 and plots were revisited in July 1996. Species cover was estimated by means of the decimal scale of Londo (1984), with 13 cover classes that were converted to mean values for data analysis. For this purpose, only the maximum species cover of

both sampling times was selected. Nomenclature follows Lambinon et al. (1998).

Parcels with a common land-use history since 1775 were delineated to determine the forest age (see Verheyen and Hermy, 2001). The forest age of parcels that have been permanently afforested since 1775 was set at 221 years. The distance of the sample points to the nearest parcel margin that has never been disturbed since 1775 was calculated, assuming that this functioned as a colonization source (sec Verheyen and Hermy, 2001).

The insolation on the forest floor was measured with a luxmeter in August 1996, at 36 points systematically distributed over each sample plot. Relative insolation of a plot was calculated as the mean value of 36-point measurements, divided by the insolation recorded in an open field.

The upper 10 cm of the mineral soil was sampled to characterize soil conditions that can determine the herbaceous vegetation. A mixture of seven samples within each $100 \, m^2$ plot was collected for analysis in August 1996. Soils were analyzed for soil moisturc, total P, plant-available P, nitrate, total N, total C and pH (KCl). It was assumed that these variables could detect both natura! gradients and anthropogenic influences on soil conditions. Total P and plant-available P were both obtained by extraction with $HClO_4$ (Olsen and Sommers, 1982) and ammonium lactate (Vanderdeelen, 1995). Both P extractions were determined according to the colorimetrie method of Scheel (1936) with molybdenum vanadate as the color reagent. For calcareous soils, the H_2SO_4 in the Scheel solution II was replaced by HNO_3 (Van Ranst et al., 1999). The relative P availability was quantified by the fraction of plant-available P to the total P content. Nitrate concentration was determined potentiometrically with a specific electrode after extraction of the fresh soil with $KAlSO_4$ solution. The modified Kjcldahl method, using Se as catalyst, was applied for the analysis of total N (Van Ranst et al., 1999). The analysis of the C content followed the Walkley and Black method and a correction factor was applied which assumed that this method analyzed 75% of the total C content (Van Ranst et al., 1999). Soil pH (KCl) was measured potentiometrically with a glass-calomel electrode in l N KCl (Van Ranst et al., 1999). The moisture content was determined gravimetrically by drying 5 g of the fresh soil for 48 h at 105 °C (Van Ranst et al., 1999).

2.3. Data analyses

Ancient-forest species were identified based on Honnay et al. (1998). Species that were not indicative for ancient forest according to Honnay et al. (1998) were subdivided into shade-tolerant and light-demanding species using Ellenberg's L index (Ellenberg, 1991). Species not listed as ancient-forest species but with an L value below 6, which indicates that they are at least tolerant to moderate levels of shadow, are further referred to as fast-colonizing woodland species. Species that are not indicative for ancient forest and with an L value equal to or above 6 are further referred to as light-demanding species. The total cover of the species groups in the sample plots was calculated as the sum of the cover of individual species.

Kendall partial rank-order correlations between environmental variables were calculated in an exploratory analysis. The impact of environmental variables on forest vegetation was investigated using direct gradient analysis (CCA) in Canoco for Windows (version 4.0). The automatic forward selection procedure for environmental variables was followed, with nine variables in the upper model: pH (KCl), forest age, distance to migration source, total N content, soil nitrate, C content, plant-available P, soil moisture, and relative insolation. Total P was not included since it was assumed that the plant-available P fraction better reflected the soil fertility level. Variables significant at the 0.05 level (Monte Carlo test) were included in the model, as prescribed by Ter Braak and Smilauer (1998). The triplots were generated in Canodraw 3.1 and Canopost for Windows 1.0.

Stepwise linear regression models were constructed to test the impact of the environmental variables on species number and total cover of each species group. The cover of competing species groups was also added to the regression models of ancient-forest species and fast-colonizing woodland species. Distance to migration source and forest age were not included in the models for light-demanding species, since it was assumed that these species are not dispersal limited, but the impact of the interaction between plant-available P and relative insolation on light-demanding species was determined. After an arcsine transformation, which has a normalizing effect on species cover data, the response of $U. \, dioica$ was

investigated in a similar manner to the total cover of the light-demanding species.

3. Results

3.1. Relations between forest age and other environmental variables

A large number of significant correlations were recorded between forest age and other environmental variables. Exceptions were the C/N ratio, soil nitrate, pH (KCI) and relative insolation (Table **1).** The C and total N contents gradually increased, from, respectively, 3.3% and 2708 rug/kg in the most recently afforested parcels to 4.3% and 3375 mg/kg in ancient forest and 19th century afforestations (Table 2). Most of the other significant correlations with the C and N contents were due to the strong correlations of both variables with forest age. Since C and N simultaneously increased, the C/N ratio was constant over all four forest-age classes. By contrast, the C/P ratio was a good indicator for the age of an afforestation (Tables 1 and 2). In spite of the increasing soil N pool

Table 1

Significance of correlations between environmental variables, tested with Kendall's tau statistic (2-sided P valuesl"

	AGE	C	CN	CP	D	M	NI	NT	PH	PL	PT
C	0.39".										
CN	0.05 NSh	0.15 NS									
CP	0_59•••	0.45".	0.12 NS								
D	-0.21*	-0.25**	0.05 NS	-0.16 NS							
M	o.5o· -	0.49***	-0.01 NS	0_44***	--0.37***						
NI	-0.09 NS	0.24**	-0.01 NS	-0.04 NS	-0.10 NS	0.12 NS					
NT	035•••	o.55**"	- -0.30***	032 ***	-0.29**	0.45· -	0.23*				
PH	-0.05 NS	-0.02 NS	0.15 NS	-0.07 NS	0.10 NS	-0.08 NS	-0.07 NS	-0.13 NS			
PL	-0.54· -	-0.23•	-0.09 NS	-0.56***	0.04 NS	-0.31•••	0.15 NS	-0.15 NS	-0.04 NS		
PT	-0_55***	–0.29"	-0.08 NS	-0.84***	0.13 NS	-0_34***	0.13 NS	-0.19*	0.06 NS	0.60***	
RI	-0.1 1 NS	-0.33***	-0.07 NS	-0.18*	0.02 NS	-0.19*	-0.26**	-O.21 -	-O.ü7 NS	0.10 NS	0.08 NS

" The following variables were investigated: forest age (AGE), C content (C), C/N ratio (CN), C/P ratio (CP), distance to migration source (0), soil moisture content (M), soil nitrate concentration (NI), total N content (NT), pH (KCI) (PH), plant-available soil P content (PL), total P content (PT) and relative insolation (RI).

h Not significant.

* p :::; 0.05.

** p :::;0.01.

••• p :::; 0.001.

Table 2

Mean values of environmental variables in four forest-age classes (years), with the standard error of each mean between brackets (abbreviations are explained in Table 1)

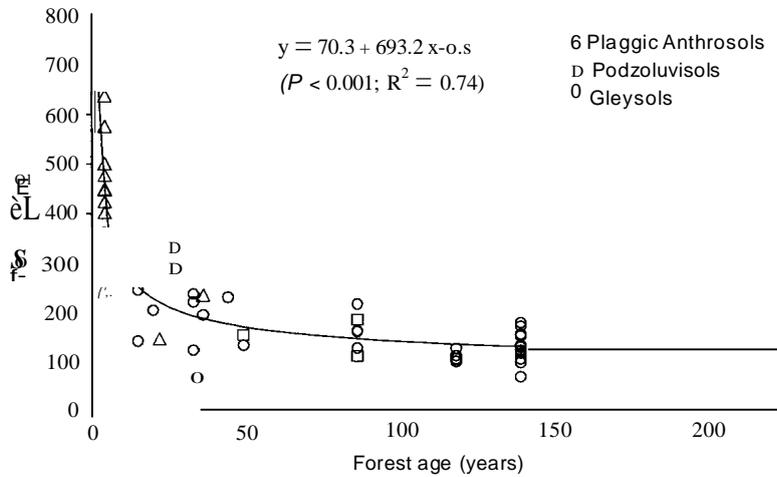|	<30	30-79	80-129	>129
C (%)	3.3 (0.2)	3.9 (0.1)	3.8 (0.2)	4.3 (0.1)
CN	12.4 (0.2)	12.0 (0.4)	11.9 (0.4)	12.7 (0.3)
CP	1 13.2 (14.9)	267.0 (49.6)	296.0 (23.2)	349.8 (22.4)
D (m)	33.2 (5.6)	16.4 (3.8)	27.1 (3.8)	18.4 (4.1)
M (%)	25.5 (1.3)	33.1 (1.9)	31.8 (1.9)	36.9 (1.0)
NI (mg/kg)	13.5 (2.3)	14.0 (3.6)	4.1 (1.1)	10.4 (2.4)
NT (mg/kg)	2708.0 (121.0)	3302.0 (179.0)	3202.0 (147.0)	3375.0 (90.0)
PH	4.4 (0.1)	4.7 (0.2)	3.9 (0.3)	5.0 (0.3)
PL (mg/kg)	89.7 (11.8)	18.3 (3.9)	17.4 (3.5)	12.1 (2.0)
PT (mg/kg)	359.0 (31.4)	175.3 (20.2)	135.5 (11.6)	128.8 (7.2)
RI (%)	19.9 (4.9)	8.7 (2.2)	12.9 (4.4)	13.8 (4.8)

Fig. 3. The total P content as a function of the forest age, with a specification of the soil type according to the Belgian soil map.

after afforestation, nitrate concentrations measured at the end of the growing season were not correlated with forest age. Nitrate values were correlated with the total N content and to relative insolation (Table 1). The negative correlation of nitrate with the relative insolation on the forest floor indicates that concentrations were relatively high in shaded forest soils (Table 1).

Both soil P fractions were negatively correlated with forest age (Table 1). Mean values of total P and plant-available P recorded in most recently afforested parcels were, respectively, three and seven times

as high as the values recorded in ancient forest and l 9th century reforestations (Table 2). In particular Plaggic Anthrosols, most of which were only afforested in 1991, are eutrophicated with P (Fig. 3); moreover an increased ratio of plant-available P to total P was determined in these soils (Fig. 4).

The positive correlation between soit moisture content and forest age (TabIe 1) is explained by the fact that waterlogged soils of the historica! core of the forest (Fig. 2) were afforested first, whereas most of the relatively dry Plaggic Anthrosols surrounding the
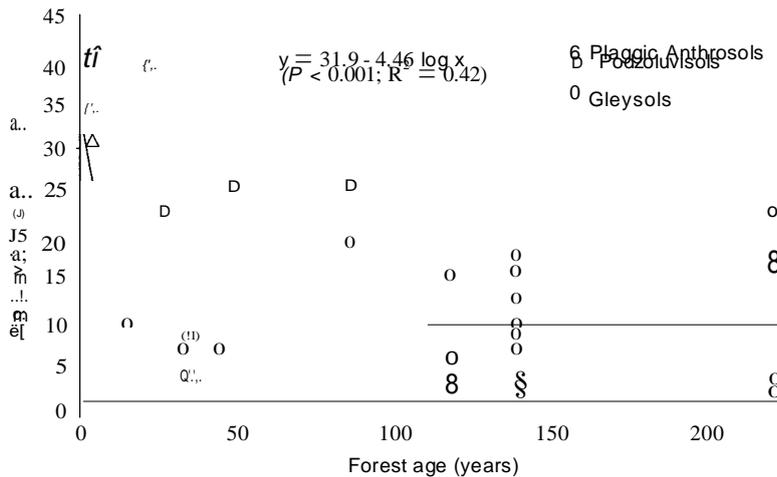

Fig. 4. The ratio of plant-available P to total P as a function of forest age, together with a specification of the soil type according to the Belgian soil map.

Gleysols were only afforested in 1991. This could also explain the observed correlations between soil moisture and soil P, soil moisture and total N content, soil moisture and C content, and soil moisture and the distance to the nearest migration source (Table 1).

The pH (KCI) value was not significantly correlated to any other variable, but the variability was higher in older forest parcels than in younger ones (Tables 1 and 2). The absence of a significant correlation between forest age and relative insolation is due to the plantation of an understorey in some of the recent afforestations.

3.2. Colonization of wood/and species in shaded and illuminated afforestations

Table 3 lists all observed herbaceous species, classified as ancient-forest species, fast-colonizing woodland species and light-demanding species with good colonization capacities. The number and total cover of ancient-forest species per plot steadily increased with increasing forest age, which is in accordance with their status (Fig. 5). The stepwise linear regression models indicated that this tendency was highly significant for the species number but somewhat weaker for the total cover (Table 4). By contrast, the number of fast-colonizing woodland species had already reached its optimum in 30-79-year-old afforestations and therefore no significant contribution of forest age was found in the regression model (Table 4). The total cover of fast-colonizing woodland species was negatively correlated with forest age (Table 4), as it had a maximum of 97% in 30-79-year-old afforestations and was lower in older forest-age classes (Fig. 5). There was no indication that forest age influenced species number and the cover of light-demanding species (Fig. 3 and Table 4). The cover of light-demanding species was lower in shaded than in illuminated sample plots, but no significant effect on the species number was detected (Fig. 5 and Table 4).

Species number and the cover of woodland species, irrespective of their colonization capacities, were higher in sample plots with a relative insolation below 8%, than in illuminated sample plots with a relative insolation equal to or higher than 8% (Fig. 5). This was observed in three of the four forest-age classes. Although no significant direct contribution of relative insolation could be assessed in the regression models on woodland species, there was an indirect effect, as reduced competition by light-demanding species had a positive influence on species number as well as the cover of ancient-forest species and fast-colonizing woodland species (Table 4).

Table 3

Classification of species using the list of ancient-woodland species of Honnay et al. (1998). in combination with Ellenberg's indicator value for insolation (L) (Ellenberg, 1991)"

Ancient! woodland species
A. maculatum (22), B. sylvaticum (6), Carex sylvatica (2), C. majalis (2), D. cespitosa (20), L. galeobdolon (l3). L. periclymenum (6), Maianthemum bifolium (2), O. acetosella (2), P. quadrifolia (14), Poa nemoralis (1), P. multiflorum (21), P. aquilinwn (2), Viola reichenbachiana + riviniana (7)

Shade-tolerant species (L < 6), not indicative for ancien! woodland
A. mosschatellina (12), Aegopodium podagraria (12), A. nemorosa (25), A. filix-femina (14), C. lutetiana (21), D. curthusiana (9), Dryopteris dilatata (8), D. filix-mas (1), Geranium robertianum (1), G. urbanum (4). M. trinervia (6), R. Jicaria (27), Stachys sylvatica (5), Stel/aria holostea (2)

Light-demanding species (L ::'.': 6), not indicative for ancient woodland
Agrostis canina (1), Agrostis stolonifera (1), Ajuga reptans (3), A. sylvestris (18), Arrhenaterum elatius (2), Cardamine flexuosa (1), Chenopodium polyspennum (1). C. arvense (9), Cirsium oleraceum (13), Cirsium palustre (6), Dactylis glomerata (1J, Elymus repens (1), Epilobium angustifolium (1), E. ciliata (IJ, E. parviflorum (1), E. tetragonum (2J, Equisetum arvense (IJ, E. cannabinum (8), · F. ulmaria (15), G. bifida + tetrahit (7), G. aparine (31), G. hederacea (19J, Glyceria fluitans (2J, Heracleum sphondylium (5), Holcus lanatus (12J, H. lupulus (8J, Hypericum dubium (IJ, /. Pseudacorus (2), Juncus effusus (2J, Listera ovata (8J, Lolium peremze (1), Lychnis flos-cuculi (1J, Lycopus europaeus (2), Mercurialis annua (1), Ornithogalum umbellatum (8), Poa trivia/is (6J, Polygonum hydropiper (3J, P. elatior (17J, Ranunculus acris (IJ, R. repen.l·(9), R. caesius (11), R. fruticosus (33), Rumex acetosa (2). Senecio vulgaris (IJ, So/anum dulcamara (2), Sonchus oleraceus (IJ, Stel/aria graminea (2), S. media (3J, Symphytum officinale (4J, Tussilago farfara (i), U. dioica (43), Valeriana repens (IJ, Vicia hirsuta (1)

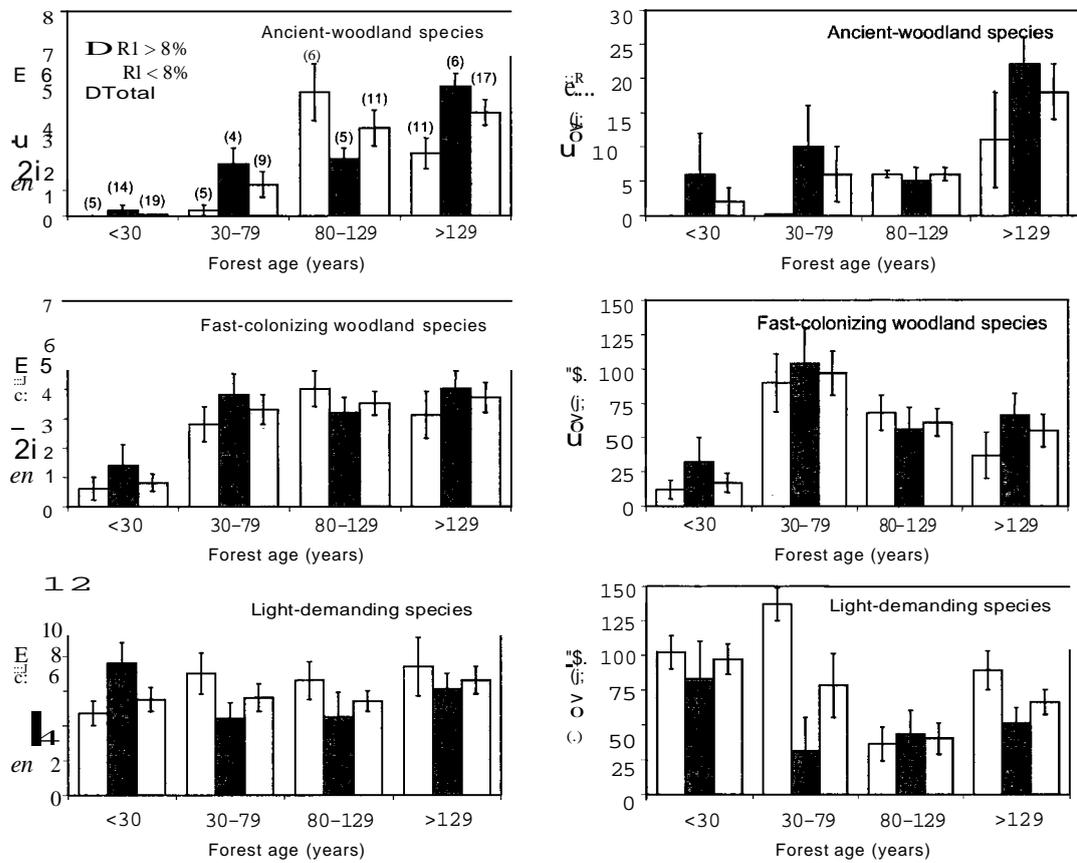"The number of observations on a total of 57 plots is stated between brackets.

Fig. 5. Comparison of mean species number and cover of the three species groups in four forest-age classes, further subdivided into illuminated sample plots (RI 2: 8%) and shaded sample plots (RI < 8%). The number of observations is stated between brackets.

3.3. Direct gradient analysis

The relation between soil types, species groups and environmental variables is visualized by two triplots (Fig. 6). The CCA indicated that plant-available P, pH (KCl), nitrate and forest age, in order of decreasing importance, were the variables that contributed significantly to the ordination model. All other variables were eliminated by the forward selection procedure. Although a strong negative correlation between forest age and total soil P content was assessed (Table 1), both variables were included. The permutation test indicated that the global species-environment relation was significant (P = 0.005). Forest age and plant-available P correlated best with the first canonical axis, pH (KCl) with the second axis and soil nitrate with the third axis. The first three axes explained 19.4% of the

variance of the species data and 89.5% of the variance of the species-environment relation.

The triplots confirm that Plaggic Anthrosols were only recently afforested and that high soil P values were determined in these soils (compare Figs. 3 and 6). Podzoluvisols are characterized by a low pH and a high nitrate concentration. Gleysols are scattered and do not occupy a specific position in the triplots.

The CCA indicated that environmental variables were also correlated with the performance of the three species groups. Ancient-forest species were found on the negative side of the first axis, corresponding with a high forest age and a low soil P content (Fig. 6). Fast-colonizing woodland species occupied an intermediate position and the group of light-demanding species with good colonization capacities had no preference towards soil P level or forest age. *Angelica sylvestris,*

Table 4
Linear regression models for species number and cover on environmental variables, and on competition by other species group (RI x PL: interaction between RI and P; CF: total cover of fast-colonizing woodland species; CL: total cover of light-demanding species; for other abbreviations see Table 1)a

Species group	R_z	F	p	AGE	M	NI	PH	PL	RI	PL x RI	CF	CL
Ancient woodland												
Species number (log)	0.70	30.0	<0.001	+++	NS	NS	++		NS	/h	NS	
Total cover (sqrt)	0.65	15.6	<0.001	+	NS	NS	+++		NS	/	NS	
Fast colonizers												
Species number (log)	0.59	18.9	<0.001	NS	+	NS	+		NS			
Total cover (sqrt)	0.52	10.9	<0.001		NS	NS	++		NS			
Light-demanding												
Species number	0.26	4.5	0.003		NS	NS	+++	NS	NS			
Total cover (sqrt)	0.40	6.8	<0.001		NS		NS	++	++			
Cover *U. dioica* (arcsine)	0.37	15.6	<Ü.001		NS	NS	NS	+++	+	NS		
Total												
Species number (log)	0.58	18.3	<0.001	+	NS	NS	++		NS			
Cover (sqrt)	0.57	11.4	<0.001		+		+++		+			

a Positive coefficient: +++ P ::; 0.001; ++ P ::; 0.01; + P ::; 0.05. Negative coefficient: −−− P ::; 0.001; −− P ::; 0.01; − P ::; 0.05.
h Not tested.

Eupatorium cannabinum, Humulus lupulus, Iris pseu-dacorus, Rubus caesius, and *R. fruticosus* were light-demanding species that preferred older forests with low soil *P* values, while *Cirsium arvense, Galeopsis bifida + tetrahit, Galium aparine, Ranunculus repens,* and *U. dioica* were indicators for recent forests on P-rich soils. The triangular pattern of the species plot confirms that soil pH had a greater influence on forest vegetation in ancient forest and the earliest afforestations than in recently afforested farmland (Fig. 6). *Arum maculatum, Brachypodium sylvaticum, Paris quadrifolia,* and *Polygonatum multiflorum* were indicators of a high soil pH, while *Lonicera periclymenum, Oxalis acetose/la, Pteridium aquilinum* and *Lamium galeobdolon* (weaker) preferred acid soils (Fig. 6).

Grasses *(B. sylvaticum* and *Deschampsia cespitosa),* fems *(Athyrium filix-femina, Dryopteris carthusiana, D. dilatata), Adoxa mosschatellina* and *Moehringia trinervia* were positively correlated with soil nitrate concentration. The ancient-forest species *L. periclymenum, Convallaria majalis, P. aquilinum* and most of the light-demanding species, which prefer a high forest age and a low soil P content, showed a negative relationship with soil nitrate concentration. Fast-colonizing woodland species were generally found on nitrate-rich soils (Fig. 6).

3.4. Linear models on species groups

The impact of soil characteristics, in combination with forest age, distance to migration source and relative insolation, was also studied using regression models (Table 4). No significant contributions were detected for total N content, C content and distance to migration source, and the variables are therefore not listed in Table 4.

Soil pH was positively correlated with the diversity of all species groups, although the correlation with fast-colonizing woodland species was relatively weak (Table 4). There was also a positive contribution to the regression model on the total cover of both woodland species groups, but not on the total cover of light-demanding species. In spite of the highly significant correlation between forest age and soil P content, both factors were included in three of the four regression models for woodland species (Table 4). These findings are in accordance with the CCA that recorded a significant effect of forest age, additional to plant-available soil P content. A high soil P content was negatively correlated with diversity and total cover of ancient-forest species and to the total cover of fast-colonizing woodland species. Competition by light-demanding species was negatively correlated with the total cover and species number of both woodland
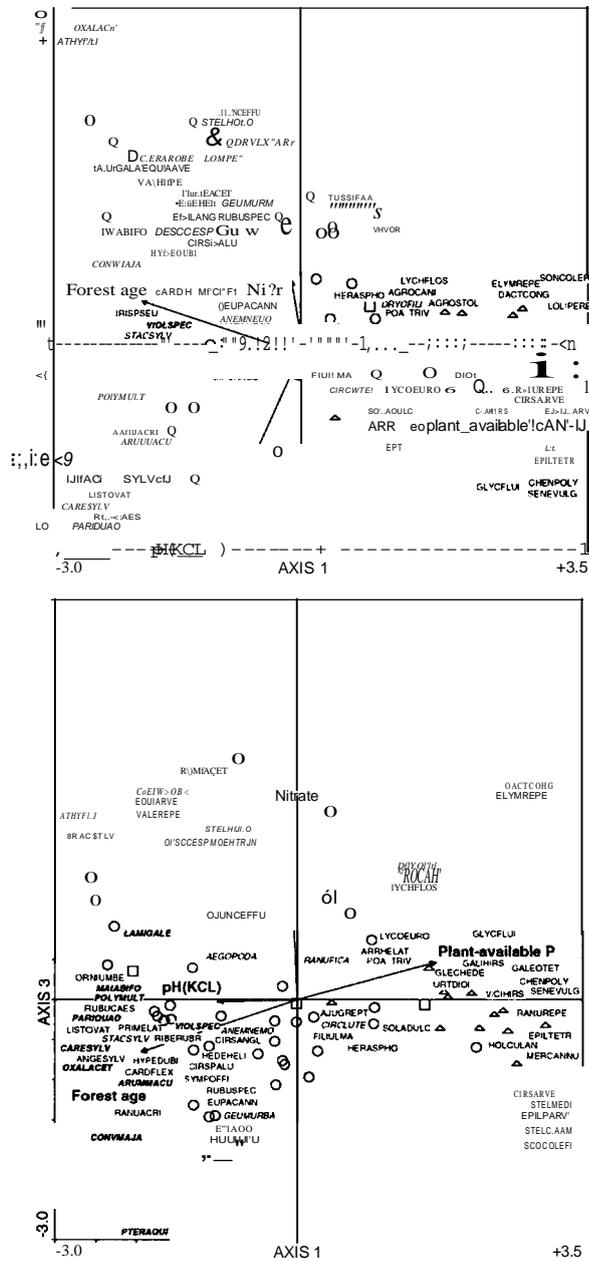
Fig. 6. Triplots of the first three axes of the CCA, both illustrating the ordination of species and sample plots in relation to four significant (P < 0.05) environmental variables. Sample plots are labeled with symbols that indicate soil types (see Fig. 3). Bold italics indicate ancient-woodland species, italics indicate fast-colonizing woodland species and the standard font type indicates light-demanding species.

species groups, irrespective of their colonization capacities (Table 4). There was no indication that ancient-forest species suffered from competitive exclusion by fast-colonizing woodland species (Table 4).

The regression model confirmed that the total cover of species with good colonization capacities and a high L indicator value, was positively influenced by relative insolation and by P enrichment. This effect of P availability was mainly due to *U. dioica,* which was dominant in well-illuminated forests on P-rich soils (Table 4). A negative interaction of relative insolation with soil P was assessed for the model on the total cover of light-demanding species and there were no indications that the combination of both factors had a stimulating effect. A negative correlation was observed between the cover of light-demanding species and the soil nitrate concentration (Table 4).

4. Discussion

4.1. Impact of land-use history on forest soil fertility

Both soil P fractions were much lower in the soils of ancient forest and 19th century reforestations than in the most recently afforested farmland soils. Several other studies report similar observations (Dzwonko 1999). The impact of agricultural land-use on soils can vary, which could explain the opposite findings of Wilson et aL (1997), who compared ancient-forest soils with soils of 100-200-year-old afforestations. Soils of secondary woodlands established on heathland contained less Ca^{2+}, Mg^{2+}, K+ and P than ancient-forest soils, but the reverse was observed in forest soils on former arable land (Wilson et aL, 1997). Froment and Thange (1967) also found soil degradation, expressed by a decrease in soil pH, as a consequence of agricultural exploitation. Agricultural crises and famine in the mid-19th century provoked a reclamation of woodland, which severely reduced the forest area in Flanders (Tack et aL, 1993). Most of the infertile and impoverished soils were abandoned in the second half of the 19th century and reforested (Tack et aL, 1993)_ A similar phenomenon was observed in Muizen forest. The total soil P contents in afforestations of the 19th and early 20th century were comparable with the P contents in ancient forests on

Gleysols and Podzoluvisols, which indicates that these soils have received little or no fertilization. By contrast, very high P contents were found in Plaggic Anthrosols that had been arable land until 1945 and then grassland until afforestation in 1991. According to the soil map, the field survey for this was performed in circa 1960, these soils are characterized by an anthropogenic Ah-horizon of at least 50 cm thickness. This observation indicates that this is old arable land, with a long history of manuring that can explain high soil P contents.

Fertilization of arable land and grassland results in an accumulation of P in the topsoil (Guggenberger et al., 1996) and as P is tightly retained in forest ecosystems (Wood et al., 1984; Binkley, 1986; Zhang and Mitchell, 1995), losses are negligible in afforested farmland. The highest total P values in Muizen forest, amounted to 600-700 mg/kg, which is comparable with values determined in a field on a calcareous clay polder soil, in Flanders (Vanderdeelen, 1995). Values recorded in ancient-forest soils and 19th century refor-estations of Muizen forest ranged between 50 and 200 mg/kg. The use of fertiliiers in previous centuries, but in particular after 1945, is deemed to be responsible for the observed accumulation of P in the upper soil of recently afforested parcels. In particular labile inor-ganic forms of P increase as a result of fertilization (Guggenberger et al., 1996). After the afforestation of farmland, P is gradually immobilized by the formation of Ca and Al phosphates. Afforestation often results in a progressive soil acidification (Goovaerts et al., 1990; Bossuyt et al., 1 999a), reducing P availability by formation of Al phosphates (Kuo, 1993). On acidic, spruce-covered, sandy soils in Denmark, most of the added fertilizer P had become immobilized 20-30 years after application (Magid, 1993). When a cal-careous field soil, which had remained uncropped for 6 years, was extracted with ammonium lactate 30% of the total P content was still recovered (Vanderdeelen, 1995). A similar level was found in the Plaggic Anthrosols of Muizen forest which had been afforested for 4 years, while no more than 20% of the total P content was extracted from Gleysols and Podzoluvisols that had been afforested for at least 100 years.

The variability in soil pH recorded in Muizen forest was correlated with forest age. Wilson et al. (1997) also found that soil variability was greater in ancient forest than in recent forests. The liming and fertiliza-tion of farmland soils could explain this low varia-bility. Fertilization with P can have a leveling effect on soil pH, since P enrichment increases soil negative charge and soil pH in acid soils (Naidu et al., 1990). As a consequence, the large pool of plant-available P can slow down acidification of recently afforested Plaggic Anthrosols towards pH (KCI) values recorded in adjacent ancient forest on Podzoluvisols, which is a reference situation.

The development of a forest soil after the afforesta-tion of farmland is expressed by a gradual increase of the topsoil organic matter content (Catt, 1994; Ver-heyen et al., 1999), a phenomenon also observed in Muizen forest. This accumulation of organic matter is accompanied by an increase in total N content (Muys et al., 1992). In spite of the influence of forest age on the total N pool and the significant effect of the total N content on nitrate concentrations, no effect of the forest age was observed on soil nitrate concentrations in Muizen forest. Although soil nitrate was not mon-itored throughout the year, but determined by a single measurement in the summer, this might indicate that there are other important factors that determine nitrate availability. The impact of forest age on nitrate avail-ability is put into perspective by the following con-siderations: (1) mean N depositions in Flanders amounted to 39 kg/ha in 1998, causing a genera) N saturation in forests (Van Gijseghem et al., 2000); (2) recently afforested farmland soils are rich in nitrate as a consequence of fcrtilization; (3) nitrification is optima) in moist soils with a muil humus (Gleysols), which is the dominant soil type in Muizen forest.

4.2. Impact of soil fertility and insolation on colonization of woodland species

Verheyen and Hermy (2001) studied the recruitment or dispersal limitation of 16 woodland species in Muizen forest. Slow-colonizing species which pre-ferred a high soil pH in our study *(P. multijlorum, P. quadrifolia , A. maculatum)* were classified as recruitment limited, while *Anemone nemorosa* and *D. cespitosa,* which were insensitive to soil pH in our study, were classified as dispersal limited. Accord-ing to Verheyen and Hermy (2001) *A. mosschatellina, Circaea lutetiana, Dyopteris carthusiana, Glechoma hederacea* and *Ranunculus ficaria* were not recruit-ment or dispersal limited. All these species, except G. *hederacea,* were classified as fast-colonizing

woodland species, with a maximal cover in afforestations with an age of 30-79 years. The relatively low cover of this species group in afforestations older than 79 years, could be due to increased competition with ancient-forest species. Verheyen and Hermy (2001) suggested that the recruitment of fast colonizers was not limited by soil pH. The relatively low variability of pH (KCl) in afforestations younger than 80 years, implies that our data is insufficient to confirm or refute this, as it probably did not cover the complete ecological amplitude of the species. However, according to Taylor and Markham (1978) and Taylor (1997), *R. ficaria* and *Geum urbanum,* respectively, can tolerate a wide range of soil pH.

Dzwonko and Loster (1997) observed a strong divergence in the development of secondary woods, as a consequence of differences in N availability and insolation that were determined by tree species. The CCA and the regression models suggest that nitrate also determined secondary succession in Muizen forest. A high cover of light-demanding species (mainly *U. dioica)* was associated with a low soil nitrate concentration and shade-tolerant, fast-colonizing woodland species preferred nitrate-rich soils. This seems to contradict Olsen (l921), who found that *U. dioica* has a high nitrate demand. Pigott and Taylor (1964) confirmed this finding but concluded that P availability, and not nitrate, is generally growth-limiting. It is assumed that consumption by light-demanding species, most of which are tall herbs, might have reduced nitrate concentrations at the end of the growing season. At sample points with a dense understorey, high nitrate concentrations were found. The combination of the high N depositions in Flanders, the presence of a clay substrate (which impaired washing out) and the relatively low productivity of the shade-tolerant herbaceous vegetation, resulted in an accumulation of nitrate. Some of the fast-colonizing woodland species found on nitrate-rich, shaded soils, are indeed known to be N indicators with a high nitrate demand. van Dobben et al. (1999) found that experimental fertilization with N resulted in an increase in *D. carthusiana* on poor sandy soils.

Koemer et al. (1997) found that several woodland species sensitive to soil degradation *(A. filix-femina, Dryopteris filix-mas, Festuca altissima, Senecio hercynicus),* occurred more frequently in former cropland and gardens with an elevated P content, than in ancient forest. In Muizen forest, woodland species sensitive to acidification might indirectly benefit from P eutrophication, as a large pool of P in Plaggic Anthrosols is a buffer against acidification towards the pH levels observed in Podzoluvisols. An introductory experiment by Verheyen (2002) showed that *Primula elatior,* an indicator of a high soil pH, also germinated abundantly on these soils and that seed limitation was the main factor limiting colonization. In spite of this potential positive effect of P eutrophication, the linear models suggested a negative influence on the diversity of woodland vegetation. A similar negative effect was found on the floristic diversity of grassland ecosystems (Janssens et al., 1998). McKendrick (l996) observed a direct negative influence of P fertilization on the germination of *Dactylorhiza fuchsii,* an orchid species that is also present on calcareous soils with low soil P values of Muizen forest. Research by Pigott (1971) demonstrated that *U. dioica* benefits from increased P availability. We assume that the negative effect of P enrichment in Muizen forest mainly is a consequence of stimulated competitive exclusion by *U.dioica.* Verheyen (2002) found that the presence of a competing vegetation cover suppressed the growth of introduced species. In this experiment, no indications were found for increased competition across the old-young forest P-gradient, but at all introduction sites a planted second layer was present which reduced light-demanding competitors.

Pigott and Taylor (l964) found that deep shadow can suppress *U. dioica* and this is confirmed by the results from Muizen forest. Low insolation reduced light-demanding species, in particular *U. dioica,* and stimulated the colonization of recent afforestations by woodland species. The understorey of poplar plantations, the dominant forest type in our research, is initially poorly developed and gradually develops with increasing forest age (Lust et al., 2001). The case study in Muizen forest indicated that the plantation of an understorey could be an appropriate management tool for stimulating the establishment of shade-tolerant woodland species.

5. Conclusion

In this case study, many soil characteristics were correlated with forest age. Forest age, soil pH, soil

P content and insolation on the forest ftoor were the most important determinants for forest vegetation. The diversity of woodland species in afforestations adjacent to ancient forest was negatively affected by light-demanding species and in particular by *U. dioica,* a species that was stimulated by a high soit P level. Two processes that are correlated with forest age can eliminate the dominance of *U. dioica* in afforestations on eutrophicated soils: the immobilization of P and the reduction of insolation on the forest ftoor. The research in Muizen forest indicated that if colonization sources are present, plantation of an understorey could reduce cover of *U. dioica* and favor the establishment of shade-tolerant woodland species. However, light-demanding species that prefer a soit with a low P availability (such as *A. sylvestris, E. cannabinum, Filipendula ulmaria* or *D. fuchsii*) do not benefit from this management. The process of P immobilization will eventually determine how long it takes for afforestations on eutrophicated farmland soils to acquire a herbaceous layer similar to that of ancient forests and l 9th century afforestations on unfertilized soils. At present, little is known about the immobilization rates in P-enriched forest soils and the possible controlling factors, such as soil texture, soil acidity, soil moisture and tree species. A combination of long-term observational studies with controlled experiments is needed to obtain more detailed information.

References

Binkley. D., 1986. Forest Nutrition and Management. Wiley, New York.

Bossuyt, B., Deckers, J., Hermy, M.. 1999a. A field methodology for assessing man-made disturbance in forest soils developed in loess. Soil Use Manage. 15, 14-20.

Bossuyt, B.. Hermy, M., Deckers, J.. 1 999b. Migration of herbaceous plant species across ancient-recent forest ecotones in centra! Belgium. J. Ecol. 87, 628-638.

Brunet, J., von Oheimb, G., 1998. Migration of vascular plants to secondary woodlands in southern Sweden. J. Ecol. 86, 429-438.

Catt, J.A., 1994. Long-term consequences of using artificial and organic fertilisers: the Rothamsted experiments. In: Foster, S., Smout, T.C. (Eds.), The History of Soils and Field Systems. Scottish Cultural Press, Aberdeen, pp. 1 19-134.

Dzwonko, Z., Gawronski, S., 1994. The role of woodland fragments, soil types and dominant species in secondary succession on the western Carpathian foothills. Vegetatio 1 1 1, 149-160.

Dzwonko, Z., Loster, S., 1997. Effects of dominant tree species and anthropogenic disturbances on species richness and ftoristic composition of secondary communities in southern Poland. J. Appl. Ecol. 34, 861-870.

Ellenberg, H., 1991. Zeigerwerte der Gefässpftanzen. Scripta Geobot. 18, 9-166.

FAO, ISSS, ISRIC, 1998. World reference base for soil resources. World Soil Resources Report, vol. 84. FAO, Rome.

Froment, A., Thange, M., 1967. Répercussion des formes anciennes d'agriculture sur les sols et la composition ftoristique. Bull. Soc. Roy. Bot. Bel. 100, 335-351.

Goovaerts, P., Frankart, R., Gérard, G., 1990. Effet de la succession de différentes affections sur les propriétés chimiques de pedons en Fagne de Chimay (Belgique). Pédologie 15, 1 79-194.

Guggenberger, G., Christensen, B.T., Rubaek, G., Zech, W., 1996. Land-use and fertilization effects on P-forms in two European soils: resin extraction and ^{31} P-NMR analysis. Eur. J. Soil Sci. 47, 605-614.

Hermy, M., 1985. Ecologie en fytosociologie van oude en jonge bossen in Binnen-Vlaanderen. Ph.D. Thesis. Ghent University.

Hermy, M., van den Bremt, P., Tack, G., 1993. Effects of site history on woodland vegetation. In: Broekmeyer, M.E.A., Vos, W., Koop, H. (Eds.), European Forest Reserves. Pudoc, Wageningen, pp. 219-231.

Hermy, M., Honnay, 0., Firbank, L., Grashof-Bokdam, C., Lawesson, J.E., 1999. An ecological comparison between ancient and other forest plant species of Europe, and the implications for forest conservation. Bio!. Conserv. 91, 9-22.

Honnay, 0., Degroote, B., Hermy, M., 1998. Ancien! forest plant species in western Belgium: a species list and possible ecological mechanisms. Bel. J. Bot. 130. 139-154.

Honnay, 0., Hermy, M., Coppin, P., 1999. Impact of habitat quality on forest plant species colonization. For. Ecol. Manage. l 15, 157-170.

Janssens, F., Peeters, A., Tallowin, J.R.B., Bakker, J.P., Bekker, R.M., Fillat, F., Oomes, M.J.M., 1998. Relationship between soil chemica! factors and grassland diversity. Plant Soil 202, 69-78.

Koerner, W., Dupouey, J.L., Dambrine, E., Benoît, M., 1997. Inftuence of past land use on the vegetation and soils of present day forest in the Vosges Mountains, France. J. Ecol. 85, 351-358.

Kuo, S., 1993. Effect of time and phospate on the growth of annual bluegrass and creeping bentgrass in two acid soils. Soil Sci. 156, 94-100.

Lambinon, J., De Langhe, J.E.. Delvosalle, L., Duvigneaud, J .. 1998. Flora van België, het Groothertogdom Luxemburg, Noord-Frankrijk en de aangrenzende gebieden (Pteridofyten en Spermatofyten). Nationale Plantentuin van België, Meise.

Londo, G., 1984. The decimal scale for relevés of permanent quadrats. In: Knapp, R. (Ed.), Sampling Methods and Taxon Analysis in Vegetation Science. Dr. W. Junk Publishers, The Hague, pp. 45-49.

Lust, N., Kongs, T., Nachtergale, L., De Keersmaeker. L., 2001. Spontaneous ingrowth of tree species in poplar poplar plantations in Flanders. Ann. For. Sci. 58, 861-868.

Magid, J., 1993. Vegetation effects on phosphorus fractions in set-aside soils. Plant Soit 149, 1 1 1-119.

Matlack, G.R., 1994. Plant species migration in a mixed-history forest landscape in eastem North America. Ecology 75, 1491-1502.

McKendrick, S.L., 1996. The effects of fertilizer and root competition on seedlings of *Orchis morio* and *Dactylorhiza fuchsiï* in chalk and clay soit. New PhytoL 134, 335-342.

Muys, B" Lust, N" Granval, Ph" 1992. Effects of grassland afforestation with different tree species on earthworm communities, litter decomposition and nutrient status. Soil Bio!. Biochem. 24, 1459-1466.

Naidu, R" Syers, J.K" Tilmann, R.W" Kirkman, J.H" 1990. Effect of liming and added phosphate on charge characteristics of acid soils. J. Soil Sci. 41, 157-164.

Oberdorfer, E., 1957. Süddeutsche Pflanzengesellschaften. Pflanzensoziologie 10. Fischer, Jena.

Olsen, C., 1921. Ecology of *Urtica dioica* L. J. Ecol. 9, 1-19.

Olsen, S.R., Sommers, L.E., 1982. Phosphorus. In: Miller, R.H., Keeney, D.R. (Eds.), Methods of Soit Analysis. Part 2. American Society of Agronomy, Madison WI, pp. 403-430.

Pigott, C.D., 1971. Analysis of the response of *Urtica dioica* to phosphate. New Phytol. 70, 953-966.

Pigott, C.D., Taylor, K., 1964. The distribution of some woodland herbs in relation to the supply of nitrogen and phosphorus in the soil. J. Ecol. 52 (SuppL), 175-185.

Scheel, K.C., 1936. Colorimetrie determination of phosphoric acid in fertilizers with the Pulfrich photometer. Z. AnaL Chem. 105, 256-269.

Tack, G., van den Bremt, P., Hermy, M., 1993. Bossen van Vlaanderen: een historische ecologie. Davidsfonds, Leuven.

Taylor, K., 1997. Biologica(flora of the British Isles, *Geum urbanum* L. J. Ecol. 85, 705-720.

Taylor, K" Markham, B" 1978. Biologica! flora of the British Isles, *Ranunculus ficaria* L. J. Ecol. 66, 1011-1031.

Ter Braak, C.J.F., Smilauer, P., 1998. CANOCO Reference Manual and User's Guide to CANOCO for Windows. Centre for Biometry, Wageningen.

van Dobben, H.F., Ter Braak, C.J.F., Dirkse, G.M.• 1999. Undergrowth as a biomonitor for deposition of nitrogen and acidity in pine forest. For. Ecol. Manage. 1 14, 83-95.

Van Gijseghem, D., De Schrijver, A., Van Hoydonck, G., Lust, N., Mensink, C., Overloop, S., 2000. Vermesting. In: Van Steertegem, M. (Ed.), Milieu- en natuurrapport voor Vlaanderen: scenarios. VMM en Garant, Leuven/Apeldoorn, pp. 367-382.

Van Ranst, E., Verloo, M., Demeyer, A., Pauwels, J.M., 1999. Manual for the Soil Chemistry and Fertility Laboratory. University of Ghent, Ghent.

Vanderdeelen, J., 1995. Phosphate immobilization in an uncropped field experiment on a calcareous soit. Plant Soit 171, 209-215.

Verheyen, K., 2002. The relative importance of seed and recruitment limitation of vascular plants in secondary forest succession. Ph.D. Thesis. University of Leuven.

Verheyen, K., Hermy, M., 2001. The relative importance of dispersal limitation of vascular plants in secondary forest succession in Muizen forest, Belgium. J. Ecol. 89, 829-840.

Verheyen, K., Bossuyt, B., Hermy, M., Tack, G., 1999. The land use history (1278-1990) of a mixed hardwood forest in centra! Belgium and its relationship with chemica! soit characteristics. J. Biogeogr. 26, 1115-1128.

Verheyen, K" Adriaens, D., Hermy, M., Deckers, S., 2001. High-resolution continuous soil classîfication using morphological soil profile descriptions. A case-study in the Muizen forest. Geoderma 101, 31-48.

Wîlson, B.R" Moffat, A.J" Nortcliff, S., 1997. The nature of three ancient woodland soils in southern England. J. Biogeogr. 24, 633-646.

Wood, T., Bormann, F.H., Voigt, G.K., 1984. Phosphorus cycling in a northem hardwood forest: biologica(and chemica! controL Science 223, 391-393.

Zhang, Y., Mitchell, M.J., 1995. Phosphorus cycling in a hardwood forest in the Adirondack Mountains, New York. Can. J. For. Res. 25. 81-87.