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# Is bark pH more important than tree species in determining the composition of nitrophytic or acidophytic lichen floras?

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*Tree species, rather than bark pH determines the occurrence of acidophytes and nitrophytes on trees.*

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ABSTRACT

To study the pH preference of epiphytic lichens, the bark pH of *Fraxinus, Tilia, Quercus and Ulmus* trees in an urban environment was measured using a flat surface electrode. The total number of trees was 253. A survey was made of the lichens in a 40 × 40 cm quadrat surrounding the pH measurement point. Our data analysis using multivariate and univariate statistical techniques indicates that the tree species is the most important factor influencing lichen colonisation, and that bark pH alone is of less importance. We hypothesize that the changed pollution climate, with strong decreases in both sulphur dioxide and ammonia concentrations over the past two decades and a concomitant general increase in bark pH, has made epiphytes less sensitive to pH.

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## 1. Introduction

A division of epiphytic lichens into acidophytes and nitrophytes is generally accepted, certain species (e.g. *Evernia prunastri*, *Hypogymnia physodes*, *H. tubulosa*) preferring a lower substrate pH than others (e.g. *Physcia tenella*, *P. adscendens*, *Xanthoria parietina*), although the term nitrophyte may suggest a response to nitrogen rather than to pH. As a consequence it has become customary to speak of acid, neutral and basic substrata. However, since the decrease of atmospheric sulphur dioxide concentration, the increase of ammonia concentration and the concomitant increase of epiphytes, acidophytic species can be seen growing among nitrophytic ones in The Netherlands. On young trees, thalli of *E. prunastri* develop closely together with *P. tenella* in a short time (Spier, 2004). Such observations suggest that the division of lichens into acidophytes and nitrophytes is less strict than often suggested in literature.

Much has been written about bark pH and other bark properties such as richness in nutrients (incl. nitrogen deposition), water holding capacity, temperature, texture, and chemistry (Barkman, 1958; Van Dobben and ter Braak, 1999; Van Herk, 2001; Van Herk et al., 2003; Davies et al., 2004; Spier, 2004; Sutton et al., 2004; Frati et al., 2006; Sparrius, 2006; Wolseley et al., 2006). pH is generally considered to be the major property of the substratum that lichens respond to. Consequently, shifts in the epiphytic lichen flora observed in polluted areas are often ascribed to a change in bark pH (e.g. Van Dobben and ter Braak, 1998; Van Herk, 2001, 2002; Van Herk et al., 2003; Marmor and Randlane, 2007), even if nitrogen is the principal pollutant.

In spite of a rather vast body of evidence for the effect of pH on epiphytic lichens, the importance of bark pH was questioned by several authors as early as the 1970s (Henssen and Jahns, 1974; Hale, 1983). More recently it has been pointed out that certain features cannot be explained by bark pH alone: Aptroot (2004) found that pH preference of *Physcia caesia* strongly depends on geographical latitude, Wolseley et al. (2006) noted that further work was necessary to understand the relationship of nitrophytes with pH, Van den Broeck et al. (2007) found some nitrophytes disappeared although bark pH increased, and Fritz (2008) stated that tree circumference is the most important of the studied variables for all species groups. Another aspect that has received little attention up to now is that pH is usually measured in water extract of bark cuttings, while lichens are only exposed to the outermost surface of the bark.

The objective of the present study was to endeavour to find an answer to the question of the importance of bark surface pH for

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epiphytes, and more specifically, if nitrophytes prefer a higher bark pH than acidophytes. Alternatively, lichen species might respond to other bark chemical or physical properties, related to e.g. the tree species or its age rather than to pH.

## 2. Material and methods

The town of Amersfoort has been used as our study area because four common wayside tree species of north-western Europe (*Fraxinus*, *Tilia*, *Quercus*, *Ulmus*) are abundant here, and carry a remarkably well-developed lichen flora consisting of both acidophytes and nitrophytes. *Fraxinus* and *Ulmus* are about 50 years old, *Tilia* and *Quercus* are about 70. The size of our study area was c. 5 $km^2$, the coordinates of its center are approx. 52°10′41′′N 5°23′07′′E. The total atmospheric deposition of nitrogen in this area was ca. 30–40 kg N $ha^{-1}$ $y^{-1}$ in 2007, $SO_2$ concentration was <4 μg $m^{-3}$, $NO_2$ concentration was c. 30 μg $m^{-3}$ and $NH_3$ concentration was <5 μg $m^{-3}$ (PBL et al., 2010). From each of 253 wayside trees lichen observations were made in a 40 × 40 cm quadrat that was located at a height of 1.50–2.00 m at different orientations. The abundance of each species was estimated in an 8-point scale comparable to Braun-Blanquet's (1964). In February and March of 2007 bark samples of about 1 $cm^2$ were collected in dry stable weather, stored in a plastic box and taken to laboratory. Immediately on arrival the samples were wetted with a few drops of distilled water and the pH of the surface was measured using an ExStik™ flat surface electrode pH meter (Kricke, 2002). Before each session the pH meter was calibrated. We allowed enough time for the pH meter reading to become stabilized (ca. 60 s). In order to estimate the within-sample variation we carried out some of the measurements in duplicate, however the variation appeared to be very small. We hypothesize that only the pH of the immediate surface is important for lichens, as water running down the tree trunk and atmospheric humidity are their main sources of nutrients (Hale, 1983; Smith, 1975). Besides pH, tree species, bole diameter and orientation were also recorded.

Both multivariate (ordination) and univariate (species-by-species) methods were used to detect the effect of the environmental variables on the epiphyte vegetation. The orientation cannot be treated as a linear variable and was therefore decomposed into two linear components, one for the North–South and one for the East–West contrast:

$$\text{N–S orientation} = \text{COS(orientation)}$$

$$\text{E–W orientation} = \text{SIN(orientation)}$$

with N–S orientation = North–South component, E–W orientation = East–West component, orientation as compass direction in degrees (0 = North, 90 = East etc.).

The aim of the statistical analysis was to detect the relation between the lichens on the one hand, and the environmental variables bark pH, bole diameter, orientation and tree species on the other hand. Tree species, which is a qualitative variable, was entered into the analysis as a separate quantitative variable for each tree species, with value 1 if a sample was on that tree species, and otherwise 0. The quantitative environmental variables were checked for normal distribution and extreme values and used untransformed. The cover codes of the species were back transformed to percentages and subsequently converted to logarithms to achieve a normal distribution.

Redundancy analysis (RDA) was used to detect the effect of the above environmental variables on the species composition of the lichen vegetation. The effect of these variables on each single species was evaluated by multiple linear regression. The multivariate analysis was carried out using the programs CANOCO (v. 4.53) and CANODRAW (v. 4.12) (ter Braak and Smilaur, 1998), for the univariate analysis GENSTAT (v. 10.2) (Payne et al., 2007) was used. The significance of the effect of each term was evaluated using the permutation test implemented in CANOCO. Results are displayed as biplots. The distinction in acidophytes and nitrophytes was taken from Van Herk (1999).

## 3. Results

The measured pH values varied between 4.07 and 6.38 (average 5.42); the average values per tree species are given in Table 1. Although most tree species have a significantly different pH, the relation between tree species and pH is not very strong (Fig. 1); 16% of the variance in pH is explained by tree species. pH increases in the order Quercus, Ulmus, Tilia, Fraxinus. Our average pH values per tree species match those given by Barkman (1958), with the exception of *Quercus*. The weak relation between bark pH and tree species allows us to separate the effects of both in a multiple regression approach. Table 2A shows the result of a forward selection of the most significant terms in RDA. All terms appear to have a significant effect except the NS component of the orientation. The effect of bark pH is

**Table 1**
Number, average diameter (in cm), and average pH per tree species, different letters indicate significant ($P < 0.05$) differences in pH.

| | *N* | Diameter | pH |
|---|---|---|---|
| Quercus | 75 | 48 ± 14b | 5.20 ± 0.44a |
| Ulmus | 47 | 53 ± 12c | 5.38 ± 0.37b |
| Tilia | 67 | 48 ± 9ab | 5.48 ± 0.36b |
| Fraxinus | 64 | 44 ± 9a | 5.65 ± 0.33c |

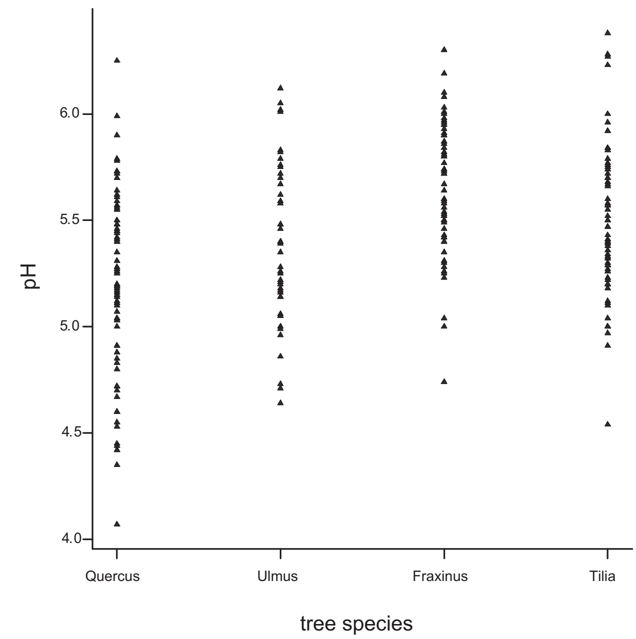


**Fig. 1.** Scatter plot of pH values per tree species.

**Table 2**
Summary of the RDA analysis. A: result of forward selection, i.e. stepwise addition of terms to the model, in each step taking the term that leads to the largest increase in percentage explained variance. *F* = increase in regression mean square on including a term, relative to the error mean square; *P* = frequency of this or a higher value of *F* under the null hypothesis in the data plus 999 random permutations. Note that the term for Tilia is non-informative after the inclusion of terms for the other three tree species and does not appear in the model. B: Top marginal variance (TMV) of each factor, i.e. the drop in explained variance on excluding this factor from the full model. Note that the sum of the top marginal variances is less than the total explained variance, the difference (in the row 'undetermined') being the portion of the variance that is not uniquely attributable to a single term. *P* = significance of the difference between a model with and without this factor determined on the basis of *F*-values by 999 random permutations.

| A | | | |
|---|---|---|---|
| Variable | *F* | *P* | %expl var |
| Quercus | 19.65 | 0.001 | 7% |
| Fraxinus | 4.49 | 0.001 | 2% |
| Ulmus | 3.02 | 0.001 | 1% |
| Bole diameter | 2.95 | 0.002 | 1% |
| E–W orientation | 2.24 | 0.010 | 1% |
| Bark pH | 2 | 0.038 | 1% |
| N–S orientation | 1.37 | 0.162 | 0% |
| SUM | | | 13% |

| B | | |
|---|---|---|
| Variable | *P* | TMV |
| Tree species | 0.001 | 7.8% |
| Bole diameter | 0.001 | 1.0% |
| Orientation | 0.01 | 1.2% |
| Bark pH | 0.023 | 0.6% |
| Undetermined | | 2.4% |

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