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Diversity and sensitivity of epiphytes to oxides of nitrogen in London

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Epiphytic biodiversity in London is related to ambient NO_x .

Abstract

This study investigated the distribution and diversity of epiphytes in London in relation to NO_x using fine-scale atmospheric dispersion modelling. The survey recorded over 3000 epiphytes from 334 trees (*Fraxinus excelsior*) representing 74 lichen, 14 moss, 7 fungal and 3 algal species. There was a significant inverse relationship between diversity and NO_x. Diversity declined where NO_x exceeded 70 μ g m⁻³ and NO₂ exceeded 40 μ g m⁻³, suggesting a phytotoxic effect. However, there was a significant positive relationship between NO_x and lichen abundance due to the ubiquitous distribution of pollution tolerant species, mainly associated with eutrophication. A scale of lichen sensitivity to NO_x has been derived.

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1. Introduction

The last major work on the lichen flora of London (Laundon, 1970) reported very low diversity attributed mainly to sensitivity to sulphur dioxide. The current study investigates the distribution and diversity of epiphytic species in London under contemporary conditions. Emissions of oxides of nitrogen (NO_x) in London have changed very little over the period, but the sources and emission heights are now quite different (GLA, 2002). Historically, NO_x was emitted at chimney height and was a product of domestic and industrial coal burning. NO_x in London is nowadays mainly emitted from vehicles at ground level where dispersion is often impeded, particularly in narrow streets and at major road junctions (Colvile et al., 2001; Arnold et al., 2005). Emissions are primarily (ca 75-95%), in the form of nitric oxide (AQEG, 2004). NO is rapidly converted to NO₂, where sufficient ozone exists, but in urban areas ozone is often relatively depleted and NO may remain

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unusually elevated (Clapp and Jenkin, 2001). Areas of highest NO_x are located at the roadside and where major transport routes converge. NO₂ is rapidly dispersed away from roads, decreasing by ca. 70% within 20–30 m of the roadside (AQEG, 2004). The ratio of NO₂ to NO_x is ca 0.25 at urban roadside, ca. 0.5 at urban background and greater in rural areas (AQEG, 2004). It is therefore very important that any study of epiphytes in relation to transport pollutants uses data appropriate to the location of the species under investigation.

London has a comprehensive air pollution monitoring network maintained to national standards under the Automated Urban Network (AUN: DEFRA, 2004). Data collected at widely spaced stations generally provide a good indication of ambient concentrations, but, owing to the rapid chemical transformations and variable dispersion characteristics across London, these measurements are not fully representative of the extent of the spatial variations experienced in this city. Measurements are therefore supplemented by data obtained from atmospheric dispersion modelling and validated against local real-time measurements.

Pollution data applied in this study were generated by Carruthers et al. (2003) using ADMS-urban (CERC, 2001),

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a computer-based model of dispersion in the atmosphere. The modelling results were validated against hourly data from 24 AUN monitoring stations in London and are generally consistent with measurements (Fig. 1) although annual mean NO_x may under-predict, particularly at high concentrations (Carruthers et al., 2003). The spatial patterns of NO_x and NO₂ in London are shown (Fig. 2). NO_x concentrations exceeded 100 µg m⁻³ throughout central London. Particulate matter in London, measured as PM₁₀, includes elemental carbon, organic compounds, CaSO₄, NaCl, NaNO₃, (NH₄)₂SO₄, Fe-rich particulates, other metals, spores, and bound water (AQEG, 2005). Annual average concentrations across the study area ranged from $24-31 \ \mu g \ m^{-3}$ (Fig. 2). Annual average concentrations of sulphur dioxide have fallen from a high of over $350 \,\mu g \,m^{-3}$ in central London in 1970 (GLA, 2002), to less than 10 μ g m⁻³ (Fig. 2). Higher SO₂ concentrations are emitted from road transport in London than nationally (GLA, 2002), with annual average SO₂ now reflecting a similar distribution pattern to other transport pollutants. Industries, mainly located to the south-east of London generate occasional short-term peak concentrations which may reach 200 μ g m⁻³ (15-min mean) in the London area (ERG, 2002) and could affect the distribution of sensitive species.

The introduction of the three-way catalyst system of motor vehicles has resulted in a significant amount of ammonia being emitted from vehicle exhausts, particularly from slow-moving traffic (Cape et al., 2004). Annual average concentrations measured at a single station in central London are consistent at ca 5 μ g m⁻³ (UK National Ammonia Network). More recent bi-monthly measurements (12 month period) at a roundabout in outer London reached 9 μ g m⁻³ and averaged 7 μ g m⁻³, with background concentrations at 3 μ g m⁻³ (James and Davies, 2005). Insufficient emissions data exist to model and analyse ammonia at a fine scale, but it is an important source of nitrogen in London and has been modelled using FRAME (Singles et al., 1998). These data were therefore included in the analysis. A small sample of climatic variables was



Fig. 1. (a) annual average NO_x(1999) from 24 Automated Urban Network Sites in London in relation to outputs from ADMS-urban (r = 0.96, p < 0.001, n = 24) (Carruthers et al., 2003). (b). NO₂ as a function of increasing NO_x(µg m⁻³), (r = 0.967, p < 0.001, n = 334).



Fig. 2. (a–c) Maps of annual average pollutant concentrations (μ g m⁻³). (a) NO₂, (b) PM₁₀, (c) SO₂. ADMS-Urban, 1999 (a,b) and 1996 (c). Sources: Carruthers et al., 2003 (a,b), Environment Agency, 2002 (c). (d) Map of 32 London Boroughs and the City of London illustrating the locations of the survey sites (Arc GIS v9).

investigated using information provided by the Meteorological Office for the years 1995 and 2000.

This study uses these data sets in order to investigate the relationship between the distribution and diversity of epiphytes on Fraxinus excelsior (European ash) in London and EU Limit Values (European Commission, 1996) for sulphur dioxide (SO_2) and oxides of nitrogen (NO_x) to protect sensitive vegetation and ecosystems. London does not exceed the Limit Value (LV) for SO_2 of 20 µg m⁻³, but exceeds the NO_x LV of 30 µg m⁻³ (GLA, 2002). Although these LVs are not applicable in large conurbations at the present time, they are nonetheless relevant to London's natural environment. The results are also discussed in relation to the potential role of lichens and bryophytes as biomonitors of transport pollution, primarily as NO_x . As far as we know this is the first semi-quantitative study of epiphytic diversity and distribution using fine-scale modelled, validated data able to represent the fast chemistry and dispersion characteristics that occur with vehicle-emitted NO_x.

2. Materials and methods

2.1. Location

London is characterised by the highest concentrations of nitrogen dioxide (NO_2) in the U.K. and transport is the major emission source (GLA, 2002). It

is therefore an appropriate location to investigate the effects of NO_x . A representative sample of London's epiphytic flora was obtained by selecting trees in public parks and open spaces in each of the 32 London Boroughs and the City of London (Fig. 2).

2.2. Survey approach and methodology

A single tree species, ash (*Fraxinus excelsior* L.), the most widely available species in the study area, was selected to reduce the influence of bark related variables (Barkman, 1958; James et al., 1977). Sites were preferentially selected where healthy, upright trees of open aspect were available in habitats corresponding primarily to urban parks and recreation areas, but in their absence country parks, woodland edge and roadside sites were also surveyed. The sizes and shapes of the parks ranged from small local parks (of about one acre or less) to large circular and oblong parks such as Regent's Park, Westminster (central London), which extends over 500 acres.

Most trees in London carry only algae; therefore, to investigate diversity it was necessary to record from trees carrying additional epiphytic flora. Trees were selected to be a minimum distance of 100 m from the road, thus the localities are termed "background" sites. The first trees meeting the selection criteria and carrying species other than algae were recorded. Records were taken from a minimum of eight trees in each borough. Frequently there were fewer than eight trees at a locality and therefore two or three different sites were surveyed within a borough before sufficient trees were found.

In total 334 trees were surveyed during 2002 and 2003. The position of each tree and associated flora was geo-referenced using a GPS (Magellan, 2002). The girth of each tree was measured at 150 cm above ground. Tree girths ranged from 20 to 350 cm with a mean of 97 cm. Girth sizes were

equally distributed across the study area and it is estimated that the average tree age was ca 30-40 years. All species on the trunk were recorded between heights of 50-150 cm above ground. Lichen and bryophyte cover was recorded within this area using a simple 3-point scale: 1 = single thallus or an area $<4 \text{ cm}^2$ for leprose/crustose species; 2 = 2-30 thalli or $<10 \times 30 \text{ cm}^2$; 3 = 30 +thalli or $>300 \text{ cm}^2$. Eight bark pH readings were taken from each tree at N, S, E, W orientations at two heights, 100 cm and 150 cm, using a flat tip electrode (BDH Gelplas double junction flat tip electrode 309/100/09 and an HI 8014 pH meter). The bark was pre-moistened with a solution of KCl (0.1 M) and the reading taken after approximately 3 min, immediately following a second application of KCl. The pH measurements ranged from pH 3.34 to 6.94. Values were converted to H⁺ ions and mean values calculated. Mean pH was 5.13. The relationship between bark pH and tree girth was investigated by grouping bark pH values into nine classes of equal girth size. Bark pH values from the north orientation were the most acidic and south orientation the least. There was a significant positive relationship between bark pH measured on the north aspect of the trunk and girth size (r = 0.911, p < 0.001, n = 9).

Thin-layer chromatography was used to identify sterile crusts and problematic lichens (Orange et al., 2001). Nomenclature follows the British Lichen Society Checklist (Coppins, 2002).

2.3. Data analysis

Survey, pollution and climatic data were converted to a format suitable for use with ARC GIS, v9 (ESRI, 1997). Tables were joined using the grid position common to each record and environmental attribute (pollution, climate, girth, bark pH) and data relevant to each species were extracted and used for the analysis.

Relationships between epiphytes and environmental variables were investigated using regression (MS Excel, Windows XP, Pearson's correlation coefficient for significance), and Canonical Correspondence Analysis (CCA), (PC-ORD v4, 2003). Canonical ordination techniques are designed to detect the patterns of variation in species data that can be explained "best" by the observed environmental variables. The resulting ordination diagram expresses not only a pattern of variation in species composition, but also the main relationships between the species and each of the environmental variables (Jongman et al., 2002).

Low species records were not included in the analysis. Co-linearity between variables was identified in a preliminary investigation and variables (mainly climatic) were removed accordingly.

3. Results and discussion

The study was carried out almost exclusively at background sites where NH_3 and NO are generally present at much lower concentrations than at roadside locations owing to their rapid conversion to NH_4^+ and NO_2 . NO_2 is therefore the major pollutant of concern, except in the City where there are no parks. Nevertheless, the study area covers a wide range of pollution concentrations from $54-213 \ \mu g \ m^{-3}$ annual average NO_x and $30-64 \ \mu g \ m^{-3}$ annual average NO_2 .

3.1. Epiphytes

All epiphytes recorded are listed in Table 1 together with lichen abundance (trees colonised × cover). Seventy-four lichen species were recorded (2998 records) from 334 trees. The mean number of species per tree was 8; highest diversity per tree was 25. The highest diversity on a single tree in any one borough was significantly correlated with total diversity within that borough (r = 0.83, p < 0.001, n = 33). The 20 most abundant lichen species given in decreasing order

of abundance were: Physcia adscendens (70% of the trees examined), Xanthoria parietina, Phaeophyscia orbicularis, Xanthoria polycarpa, Physcia tenella, Amandinea punctata, Parmelia sulcata, Lecanora expallens, Melanelia subaurifera, Xanthoria candelaria, Candelariella reflexa, C. vitellina, Lecanora chlarotera, L. dispersa, Scoliciosporum chlorococcum, Lecidella eleaochroma, Evernia prunastri, Lepraria incana and Ramalina farinacea. This assemblage is dominated by pollution tolerant (Seaward, 1997) and nitrogen tolerant ("nitrophyte") species (Barkman, 1958; James et al., 1977; van Herk, 2002; Seaward and Coppins, 2004). E. prunastri and L. incana are believed to be associated with acidity (AIW scale, van Herk, 2002). Many of these species remained in London during the highest SO₂ and NO_x "regimes", but then were largely confined to stone (Laundon, 1970), suggesting that substratum pH was an ameliorating factor. Further evidence that substratum pH plays a major role in lichen colonisation has since been reported (Seaward and Coppins, 2004; Wolseley et al., 2005). Green algae were present in all lichens recorded and where identified, as Trebouxia spp.

Orthotrichum diaphanum was the most widely distributed bryophyte and was recorded at annual average NO_x concentrations between 54 and 168 µg m⁻³. Eurhynchium praelongum was recorded from seven trees (max. NO_x 72 µg m⁻³), but confined to the north of London. These two bryophytes are included with lichens in all further data analysis. Other bryophytes (Table 1) were recorded only where NO_x concentrations were below 68 µg m⁻³ (possibly lower because they were woodland edge species), with the exception of *Dicranoweisia cirrata* (maximum concentration 98 µg m⁻³) suggesting a high sensitivity to contemporary conditions, although climatic factors could also be highly relevant (Bates et al., 2004).

Three free-living algae were identified: *Apatacoccus* spp., *Prasiola crispa*, *Trentepohlia* spp. Six non-lichenised fungi were recorded (Table 1).

3.2. Data analysis

The CCA analysis identified transport related pollutants NO₂, PM₁₀, NO_x and SO₂ as being the strongest contributors to the first ordination axis (Fig. 3) (Eigenvalue 0.109). The standardised coefficients were: -0.52, -0.097, -0.035 and -0.024, respectively. Species associated with these pollutants in the CCA were generally also the most abundant, pollution tolerant species and mainly associated with nutrient rich environments in the literature (Davies, 2005). Other important environmental factors affecting species distribution were identified as minimum temperature (-0.306), bark pH (0.299), rainfall (0.384) and girth (-0.137). The CCA results support the hypothesis that the distribution of some species is associated with transport, but also identify other important environmental variables that are known to affect distribution.

3.2.1. NO_x

 NO_x concentrations were grouped into classes of equal size (10 $\mu g\ m^{-3}$ intervals) and plotted against the total number of

Table 1 Species recorded from 334 Fraxinus excelsior in London (2002–2003)

	LICHENS	LICHENS	LICHENS	LICHENS	BRYOPHYTES	LIVERWORTS
Order: Family:	Arthoniales Arthoniaceae	Lecanorales Acarosporaceae	Lecanorales Micareaceae	Teloschistales Teloschistaceae	Amblystegium serpens Brachythecium	Frullania dilatata Lophocolea bidentata
	Arthonia radiata (1) A. spadicea (1)	Strangospora pinicola (15)	Micarea prasina	Caloplaca flavocitrina (2) Xanthoria candelaria (235)	rutabulum Dicranoweisia cirrata Dicranum scoparium	NON-LICHENISED FUNGI
		Bacidiaceae	Parmeliaceae	X. parietina (415)	Eurhynchium praelongum	Athelia arachnoidea
	Chrysothricaceae Chrysothrix flavovirens (2) Rocellaceae Schismatomma decolorans (1)	Bacidia arceutina (1) B. delicata (51) B. friesiana (1) B. laurocerasi (1) Cliostomum griffithii (2) Lecania cyrtella (30)	Evernia prunastri (94) Flavoparmelia caperata (62) F. soredians (22) Hypogymnia physodes (30) H. tubulosa (1) Hypotrachyna revoluta (4)	X. polycarpa (410)	Grimmia pulvinata Hypnum andoi H. cupressiforme H. resupinatum Orthotrichum affine O. diaphanum	Gloniopsis praelonga Hysterium pulicaris Ionotus hispidus Lachnella alboviolascens Lichenoccus erodens
	Pyrenulales <i>Monoblastiaceae</i> Anisomeridium biforme (8)	L. naegelii (1) Candelariaceae Candelaria concolor (21) Candelariella reflexa (188)	Melanelia exasperatula (2) M. glabratula (57) M. subaurifera (211) Parmelia sulcata (284) Parmotrema chinense (34)	Incertae sedis Trichotheliales <i>Trichothliaceae</i>	O. lyellii Rhynchostegum confertum Ulota crispa	ALGAE Apatacoccus spp. Prasiola crispa Trentepohlia spp.
	Gyalectales <i>Gyalectaceae</i> <i>Dimerella pineti (21)</i>	C. vitellina (144) C. xanthostigma (3) Catillariaceae Catillaria chalybeia (1)	Punctelia subrudecta (79) P. ulophylla (16) Usnea cornuta (4) Phlyctidaceae Bhurtic groups (24)	Porina chlorotica (3) Anamorphic Lepraria incana (102) L. lobificans (2)		
		Cladoniacoao	Phlychs argena (24)			
		Cladonia conjocrava (4)	Physciaceae			
		C. fimbriata (4)	Amandinea punctata (335) Diploicia canescens (18)			
		Lecanoraceae Lecanora albella (2) L. barkmaniana (2)	Hyperphyscia adglutinata (39) Phaeophyscia orbicularis (438) P. nigricans (20)			
		L. carpinea (38) L. chlarotera (128)	Physcia adscendens (548) P. aipolia (19) P. annin (45)			
		L. compatiens (5) L. confusa (45) L. conizaeoides (30)	P. caesta (45) P. dubia (31) P. tenella (406)			
		L. dispersa agg (143). L. armallans (200)	Physconia grisea (36)			
		L. expanens (299) L. muralis (5) L. persimilis (4)	R. gennarii (26) R. subexigua (1)			
		L. symmicta (79) Lecidella elaeochroma (134) L. scabra (10)	Ramalinaceae Ramalina farinacea (69)			
		Scoliciosporum chlorococcum (154)				



Fig. 3. CCA results. Lichen and bryophyte distribution in relation to environmental variables. Nitrophytes are largely associated with transport pollutants and acidophytes with bark pH (designation after van Herk, 2002).

species (lichens and two mosses, 76 species) in each class and the number of first records (the first time a species was recorded) in each class (Fig. 4). There was a significant inverse relationship between NO_x and diversity (r = 0.80, p < 0.001, n = 15) and first records within each class (r = 0.85, p < 0.05, n = 6), (Fig. 4). Diversity was highest (60 species) in the first two classes $(50-69 \ \mu g \ m^{-3})$ and then declined with just fourteen species remaining above 200 μ g m⁻³. First records were also highest at lowest concentrations, declining to 3-5 species in each class up to concentrations of $110 \ \mu g \ m^{-3}$, after which there were no further first records. There was a significant positive relationship (r = 0.85, p < 0.001, n = 76) between abundance (number of trees colonised \times cover) and the highest concentration at which species were recorded (Fig. 4), showing that the most NO_x tolerant species are widely distributed and abundant. New records and rare species were generally confined to areas of low NO_x where the ratio of NO to NO_x is generally at its lowest (Fig. 1). However, there were a few notable exceptions. Species tolerant of NO_x, but with low abundance and not recorded in areas of low NO_x were: *Phaeophyscia nigricans*, *Rinodina gennarii*, R. subexigua, Lecanora barkmaniana and L. compallens. The latter two were recently described from the Netherlands and associated with roadside trees (Aptroot and van Herk, 1999; van Herk and Aptroot, 1999). R. gennarii is designated

a "nitrophyte" in the Dutch scale for ammonia (van Herk, 2002). *Rinodina exigua* was also recorded with a high mean NO_x value. These results suggest these species may be stimulated by high NO_x concentrations. However, such environments are generally also characterised by high levels of ammonia and particulate matter (dusts).

3.2.2. NO₂

Species were grouped into classes of equal size (5 μ g m⁻³ intervals) and plotted against the number of species (lichens and two mosses) in each class and the number of first records in each class (Fig. 4). Biodiversity was highest (59 species) in the second class (35–39 μ g m⁻³) and then declined with seventeen species remaining in the highest class (60–64 μ g m⁻³). There was a significant inverse relationship between NO₂ and first records (r = 0.96, p < 0.01, n = 5), declining to just one (*Phaeophyscia nigricans*) between 50 and 54 μ g m⁻³. Species associated with high NO₂ were generally those associated with high NO_x concentrations with the exception of *Flavoparmelia caperata*.

3.2.3. PM₁₀

Species found in areas of high PM_{10} are generally the same pollution tolerant species associated with NO_x and SO₂, with the exception of *Phlyctis argena* and *Micarea prasina*. The



Fig. 4. (a) Diversity and first records as a function of NO_x (μ g m⁻³) (Diversity: r = 0.8, p < 0.001, n = 15; First records: r = 0.85, p < 0.05, n = 6); (b) Lichen abundance (trees colonised × cover) and maximum concentrations of NO_x (μ g m⁻³). Species recorded at concentrations above 200 μ g m⁻³ from left to right are: *Physcia nigricans, Lepraria incana, Scoliciosporum chlorococcum, Lecanora dispersa, Melanelia subaurifera, Xanthoria candelaria, Parmelia sulcata, Lecanora expallens, Amandinea punctata, Physica tenella, Xanthoria polycarpa, X. parietina, Physcia orbicularis, P. adscendens (r = 0.85, p < 0.001, n = 76). (c) Biodiversity and first records as a function of increasing NO₂ (\mug m⁻³) (First records: r = 0.96, p < 0.01, n = 5).*

latter is now abundant at roadside sites of medium traffic flow (James and Davies, 2005) and is also associated with pollution, although not specifically N or S compounds (Dobson, 2004).

3.2.4. Minimum temperature and rainfall

Lichen sensitivity to urban environments is well documented (Coppins, 1973). Temperatures in London remain 2-3 °C warmer in the city centre and inner suburbs throughout the year, although temperatures in central London occasionally fall below zero. Annual average relative humidity in London (measured at London Heathrow) varied between 73% and 82% over a 27-year period (D.R. Middleton, pers. commun.), slightly below that suggested for optimum lichen metabolism for green algae (Nash, 1997), and in central London averaged 62% during August and September 2004 (A. Hudson-Smith, pers. commun.). Rainfall fluctuates year on year, but is lower in the centre of London. Species associated with areas of high rainfall were Dimerella pineti, Lecidella scabra and the moss Eurhynchium praelongum. Species associated with low rainfall and minimum temperature were generally the most pollution tolerant (Fig. 3).

3.2.5. Acid bark and girth

The CCA analysis identified a positive relationship between the following species and acid bark: Anisomeridium biforme, Strangospora pinicola, Hypogymnia physodes, Evernia prunastri, Ramalina farinacea, Lepraria incana and Parmotrema chinense. Most are classified as acidophytes (van Herk, 2002) and were not associated with transport pollution.

Records of acidophytes (Hawksworth and Rose, 1970; van Herk, 2002) have continued to decline (Bates et al., 2001) and for some species distribution was relatively low (abundance in brackets) (Fig. 5): A. *Lecanora conizaeoides* (30), B. *Hypogymnia physodes* (30), but not for all: C. *Evernia prunastri* (94), D. *Lepraria incana* (102).



Fig. 5. Distribution of species associated with acidity: (a). *Lecanora conizaeoides* and (b). *Hypogymnia physodes* (both in decline); (c) *Evernia prunastri*and (d) *Lepraria incana* (both widely distributed).

Large girth was important in the second CCA ordination axis for *Strangospora pinicola*, *Physconia grisea*, *Physcia caesia* and *P. dubia*, species associated with eutrophication and nutrient-rich dusts (Gilbert, 1976; James et al., 1977). *Lecanora carpinea*, *L. confusa* and *L. symmicta* were widely recorded *and* demonstrated a preference for trees with a small girth. These species are more usually confined to twigs, which were not sampled in this study.

3.3. Diversity and EU limit values

The critical levels to protect sensitive vegetation and ecosystems were derived mainly from studies carried out on higher plants and focus on the effects of NO₂ (WHO, 2000). More recent studies investigate a wider range of vegetation and the impact of other N compounds as well as transport emissions generally (NEGTAP, 2001; Bell and Treshow, 2002; Bignal et al., 2004). Many different approaches have been applied to investigate the impact of NOx in both field studies and chamber fumigation studies (Fuentes and Rowe, 1998; Bell and Treshow, 2002; Gombert et al., 2002; Batty et al., 2003; Lorenzini et al., 2003; Purvis et al., 2003, 2004). Fumigation studies on higher plants using single and mixed transport emissions have identified direct toxicity effects, enhanced growth and element accumulation with varying degrees of sensitivity for different species (NEGTAP, 2001; Bignal et al., 2004). Nitrogen-compounds also contribute to acidification and eutrophication (NEGTAP, 2001). Some studies suggest that exposure to motor vehicle exhaust has little additional influence compared with exposure to NO_x alone (Bignal et al., 2004).

NO has been recognised as having quite distinct effects from those attributed to NO_2 and may be the more toxic component of NO_x (Wellburn, 1990; Mansfield, 2002). It is also an important signalling mechanism in animal cell function (Mansfield, 2002) and in plant defence mechanisms.

Effects of NO₂ on lichen physiology were first demonstrated at concentrations of 3760 μ g m⁻³ and 7562 μ g m⁻³ under laboratory conditions (Nash, 1976). Many studies show that traffic emissions affect lichens from the cellular to community level, although the factors responsible are poorly understood (Nash, 1988; Von Arb and Brunold, 1990; Seaward and Coppins, 2004). Angold (1997) reported the loss of terricolous lichens near roads in the New Forest and suggested that competition from faster growing species was a contributory factor. Nitrogen concentrations in Physcia adscendens growing adjacent to roads were found to increase with proximity to roads of high traffic flow, but not in *Hypogymnia physodes* (Gombert et al., 2002). NO_x was considered a possible factor preventing colonisation of Parmelia saxatilis adjacent to busy roads in central London (Batty et al., 2003). Traffic flow rates were linked with dusts and enhanced nitrogen and zinc levels in Parmelia sulcata and identified as a factor contributing to its poor health in Burnham Beeches, west of London (Purvis et al., 2003, 2004). NO₂ derived from traffic emissions limited lichen diversity (expressed as an Index of Atmospheric Purity "IAP") in Seville, Spain (Fuentes and Rowe, 1998) and in Tuscany, Italy (expressed as a lichen biodiversity value,

Lorenzini et al., 2003). NO_x derived from domestic heating emissions was considered responsible for changes in diversity at Pistoia, Italy (Loppi and Corsini, 2003).

Further evidence of varying sensitivity to transport emissions and NO_x was found in *Xanthoria parietina* which was unharmed following exposure to high traffic emissions, unlike the more sensitive *Ramalina duriaei*, which showed extensive physiological changes (Silberstein et al., 1996a,b). A superior response by *X. parietina* to oxidative stress was identified as a key factor. Other studies have reported antioxidant responses in lichens following exposure to NO_x, SO₂ and ozone (Bates, 2002; Cuny et al., 2002).

There is much empirical evidence from field studies linking many of the pollution tolerant lichens to nitrogen (Barkman, 1958; James et al., 1977; Seaward and Coppins, 2004; Davies, 2005). Laboratory-based studies have recorded high N accumulation within the thalli of many species (Purvis et al., 2003, 2004; Gaio-Oliveira et al., 2004; Gombert et al., 2002). These associations led to the term "nitrophyte". A scale of sensitivity to ammonia was devised, although it was later found that bark pH was the main factor affecting changes in community structure on oak (van Herk, 2002). Conversely, some species are highly sensitive to N and prefer a more acidic environment. High deposition of nitrogen can be directly toxic to terricolous lichens and mosses of heathland areas and some epiphytes (Pitcairn et al., 1991; van Herk, 2001; Massara, 2004).

Whilst the focus of this study is NO_x , it is recognised that many other pollutants are associated with transport. In particular ammonia and ammonium compounds as well as dusts, heavy metals, VOCs and PAHs, may be contributing to the results. In addition, the modelled data, although validated against actual measurements, may occasionally over- or under-estimate NO_x. Furthermore, urban environments are less humid and warmer than rural areas which will restrict distribution. In addition, recording was confined to *Fraxinus excelsior*, a neutral bark tree species. These factors must be taken into consideration when summarising the results of this study, as well as the avoidance of trees without epiphytes.

Lichen diversity in London has increased by an order of magnitude since the 1970s (Laundon, 1970) (Fig. 6). Much can be attributed to reductions in SO₂ and acidity (Hawksworth, 2002), but this study shows that the number of lichens in many of the central London Boroughs is now significantly higher than in some outer London Boroughs (Fig. 6). This is largely explained by the distribution of the pollution tolerant species, many of which are also associated with eutrophication. Few rare species were found; some not seen in London for decades have returned and others are new records for the Capital (Waterfield, 2002). In areas of highest NO_x, species recorded belong almost exclusively to the families Candelariaceae, Physciaceae and Teloschistaceae. Further evidence of the unremarkable flora is provided by the presence of just two fruticose species, Ramalina farinacea and Usnea cornuta, the latter with a thallus length not exceeding 1.5 cm.

Three of the most widely distributed species, *Xanthoria candelaria* group, *X. parietina* and *X. polycarpa*, are characterised by the presence of parietin, an antioxidant anthraquinone (Søchting and Lutzoni, 2003), which is the pigment that



Fig. 6. (a). Lichen diversity on trees, 1970. (b) Lichen diversity on Fraxinus excelsior, 2004.

accounts for the usually bright yellow-orange thallus in the family *Teloschistaceae*. Many samples of *Xanthoria parietina* and the *X. candelaria* group recorded from trees in open aspects in London were grey, not yellow, suggesting that parietin was depleted. Further investigation (E. Haden, pers. commun.) found that the orange colour of *X. parietina* was inversely related to NO_x which supports the hypothesis that less parietin is produced in response to increased NOx concentrations. Antioxidants can neutralise free radicals and mitigate the impact of pollution (Silberstein et al., 1996a,b; Cuny et al., 2002; Kelly, 2003) and may therefore partly explain the abundance of these pollution tolerant species.

A tentative scale of lichen sensitivity to NO_x (and associated transport pollutants in urban areas) on neutral to eutrophicated bark has been deduced using the mean and maximum tolerance exhibited by the species recorded in this study (Table 2).

Table 2

Tentative scale of sensitivity (1 = tolerant, 10 = sensitive) to NO_x for lichens growing on trees with neutral bark (*Fraxinus excelsior*)

	Mean NO _x	Peak NO _x	Rank
Lecanora dispersa, Phaeophyscia orbicularis, Physcia nigricans,	100	>200	1
Scoliciosporum chlorococcum			
Amandinea punctata, Physcia adscendens,	90	>200	2
P. tenella, Xanthoria parietina, X. polycarpa			
Lecanora expallens Lepraria incana	75	200	3
Melanelia subaurifera, Parmelia sulcata, Rinodina gennarii,	15	200	5
Xanthoria candelaria			
Candelariella vitellina, Flavoparmelia	90	160	4
caperata, Lecidella eleaochroma,			
Orthotrichum diaphanum ^a , Physcia			
aipolia, Punctelia subrudecta,			
Schismatomma decolorans			
Candelariella reflexa Lecanora	85	130	5
barkmaniana, L. albella L. chlarotera,			
L. compallens, L. conizaeoides,			
Micarea prasina, Phlyctis argena,			
Rinodina exigua, R. subexigua			
Anisomeridium biforme, Bacidia delicata,	82	120	6
Evernia prunastri, Lecanora confusa,			
Punctelia ulophylla Ramalina farinacea			
Candelaria concolor, Diploicia canescens,	80	100	7
Flavoparmelia soredians, Hypogymnia			
physodes, Lecanora carpinea,			
L. muralis, L. symmicta, Physcia caesia,			
Physconia grisea			
Lecania cyrtella, Parmotrema chinense,	75	95	8
Physcia dubia, Strangospora			
pinicola, Usnea cornuta			
Arthonia radiata, Cliostomum griffithii,	65	75	9
Dimerella pineti, Eurhynchium			
praelongum", Hyperphyscia adglutinata,			
Hypotrachyna revoluta, Lecanora			
persimilis, Lecidella scabra, Lepraria			
iobificans, Melanelia exasperatula,			
Melanella glabratula	50	= =	10
rorina chiorotica	50	22	10

Ranks are calculated from annual average NO_x and highest NO_x concentrations (modelled data using ADMS-urban provided by CERC, 2003). ^a Moss. The product of the highest concentration and the mean concentration calculated over the sites occupied by each epiphyte has been used to rank them.

4. Conclusions

The diversity of epiphytic species in London has continued to increase from the low experienced in the 1970s with over 100 species recorded from *Fraxinus excelsior* alone. Some interesting species were recorded where NO_x was lowest, but there appears to be a phytotoxic effect where NO_x exceeds 70 µg m⁻³ and NO_2 exceeds 40 µg m⁻³. All areas surveyed exceed the EU Limit Value for NO_x for sensitive vegetation and ecosystems of 30 µg m⁻³. Species present in areas of highest NO_x are generally considered pollution tolerant and are mainly associated with eutrophication. Further survey and fumigation studies are required to develop the NO_x scale for biological monitoring and conservation purposes.

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