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Modelled and observed surface soil pollen deposition distance curves for isolated trees of *Carpinus betulus, Cedrus atlantica, Juglans nigra* and *Platanus acerifolia.*

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Abstract

Source-distance relationships for pollen deposited directly into surface soil have been rarely undertaken, particularly for a single or isolated source, rather than a forest, grove or plantation. This study aimed to determine surface soil pollen deposition patterns from single, isolated source trees and to compare the results to Gaussian model curves for the same trees.

Four isolated tree pollen sources were chosen in Worcester, UK: *Carpinus betulus, Cedrus atlantica, Juglans nigra* and *Platanus acerifolia.* Surface soil *s*amples were collected at 1, 5 and then every 10 metres, up to 100 metres distance from the main trunk of each source along the prevailing wind direction during flowering. A Gaussian dispersion model was used to estimated source strength using tree height and width and wind speeds on days when flowering was occurring and when the wind direction flowed along the sampling transect. This model simulated the expected concentration and deposition along the sampling transect.

Modelled and observed results showed that most pollen was deposited beneath the canopy (range: 63% – 94%) in an exponentially decreasing curve and the tailing off started from around the outer edge of the canopy in most cases. The amount of pollen deposited at 50 metres was no more than 2.6% of total deposition in the samples for any tree and at 100m no more than 0.2%. Tree height, width and wind speed during the pollination period were found to be the main parameters affecting deposition away from the source.

Key words: forensic palynology, aerobiology, source-distance relationship

Introduction

This research has been conducted partly with the aim of increasing the empirical research knowledge base for forensic palynology but may also be of interest in aerobiology. In crime cases, pollen concentrations found in soil on an exhibit can provide information about the vegetation of a possible crime scene, or help link a suspect to one, and it is usually the top few millimetres of soil that are of interest (Milne et al. 2005; Adams-Groom 2012). Surface soil is therefore an important substrate to focus research on in this context. In some samples, the abundance of a particular taxon found in soil on an exhibit suggests that the source plant was nearby but quantifying this distance remains difficult. For many insect pollinated plants, it is already known that the presence of their pollen tends to indicate that the source plant is at the scene, since the pollen is usually heavy and/or sticky and unlikely to become airborne in significant quantities (Milne et al. 2005). For trees, however, the height, exposure to the wind, pollination strategy and other factors can all increase the chances of extended pollen dispersal, as well as affecting the local deposition. This type of information, however, is scarce and the escape fraction from local sources has been highlighted as an important knowledge gap in aerobiology (Sofiev & Bergmann 2013), thereby underlining the connection between palynology and aerobiology in this context.

In the UK, *Betula, Pinus, Alnus* and *Quercus* are the most frequent and abundant tree taxa pollen found in soil samples (Brostrom et al. 2008; Adams-Groom 2015). The trees themselves are very common in the UK (Forestry Commission, 2001), as well as much of Europe (Skjøth et al. 2008), are wind-pollinated and produce plenty of pollen (Adams-Groom et al. 2002; Skjøth et al. 2015). Certain other tree pollen types, such as *Platanus* for example, have limited distribution and lower abundance (e.g. Bricchi et al. 2000) and are usually only found in larger numbers in urban/suburban areas (Konijnenedijk et al. 2005). Such taxa are therefore of greater interest to the forensic palynologist, including the subjects in this particular study, *Juglans nigra, Carpinus betulus, Cedrus atlantica* and *Platanus acerifolia* (Adams-Groom 2015).

Released pollen is subject to a number of variables before and during deposition, such as pollen weight, settling speed, impaction, turbulence, wind-speed and air convection currents (Di-Giovanni et al. 1989; McKibbin 2006). Once pollen has deposited on the ground or other surfaces, it is then subject to further processes to transfer it from the surface to within the soil, particularly by rain wash and earthworms (Faegri and Iversen 1992; Davidson et al. 1999). Although soil samples contain more than one season's pollen, observational studies often use moss pollsters where the type of accumulation seen in soil is absent, or Tauber traps which are less affected by soil processes such as faunal and fungal action (Lisitsyna and Hicks 2014).

It has been shown that isolated smaller sources, such as *Ambrosia* or *Platanus*, can have a large impact on locally observed pollen concentrations if the sources are sufficiently close to the monitoring station (Sommer et al. 2015; Bricchi et al. 2000). Local scale models can describe how allergenic tree pollen types should theoretically be dispersed on the local scale e.g. Sugita (1994). Generally, they show an exponentially decreasing curve with a long tail where the shape of the curve away from the source mainly depends on the release height (Skjøth et al. 2013). Hofmann et al. (2014) found a power model best described the dispersion of maize pollen, stating that exponential models underestimate exposure for distances greater than 10m. Other common tools used with pollen and spores are various types of the Gaussian dispersion model. They can incorporate physical processes relating to dispersion (Van Leuken et al. 2016) and are substantially simpler and less time consuming to use compared to more advanced models such as Computational Fluid Dynamic (CFD) models (Van Leuken et al. 2016).

Existing research into the source-distance relationships of pollen from trees includes forest deposition studies which predict and/or observe an exponential decrease in pollen deposition away from a large source, either within the forest or away from its margins (Di-Giovanni et al. 1989; Brown 1999; Bunting 2002). Most observational studies of pollen deposition into surface soil, with distance from a known source, concern experimental plots of maize, grass or ragweed, as discussed by Sofiev and Bergmann (2013) and Hofmann et al. (2014). Turner and Brown (2004) looked at *Vitis* (Vine) pollen, also demonstrating an exponential decline in surface soil pollen. Articles describing observational deposition from group sources of a single tree type include the following: Anderson (1990) examined the dispersal of *Sequoidendron giganteum* pollen beyond groves of this taxon and found that most pollen was deposited within 100m of the source, largely flat-lining thereafter up to 5000m. Bricchi et al. (2000), studied pollen deposition over distance for a group of sixty *Platanus* trees, demonstrating that approximately 25% of the pollen fell within 400m of the source and at 2750 m only 9 grains were found. No articles were found by the authors that focussed purely on surface soil samples along a line transect from single or very small groups of trees, despite the importance for both palynology and aerobiology.

This research aimed to determine surface soil pollen deposition patterns, using standard palynological approaches, from single, isolated tree sources and to compare the results to simulated Gaussian model curves produced for the same trees.

Materials and Methods

Selection of trees

The main criteria for assessment of pollen concentrations in surface soil with increasing distance from the tree source, was to find isolated trees, either as individuals or very small groups, so that the footprint on the local scale was well-defined and related to a single source, e.g. Michel et al. (2010). The second criteria was for the sampling surface to have been consistently managed over the previous few years. Finally, the trees needed to stand where an open sampling distance of 100m could be accessed along the prevailing wind direction of the previous pollination period (Michel et al. 2010, 2012). In addition, it was also desirable to look at trees that are not common or widespread in the UK, ensuring that observed pollen can be attributed to only localized sources (Sommer et al. 2015). All of these conditions were met on the University Of Worcester campus and four tree sources were selected: *Carpinus betulus* (three trees in a line, sampling from centre tree), *Cedrus* *atlantica* (single), *Platanus acerifolia* (pair, sampling from base of one) and *Juglans nigra* (single). For comparison, *Pinus* pollen was counted within the *Carpinus betulus* transect. *Pinus nigra* trees occur beside the *Carpinus* but there are other pine trees scattered in the vicinity too. All the trees were mature and had been observed flowering for at least five years.

The sampled surfaces mostly comprised open areas of regularly mown lawn. However, under the *Platanus* tree an area of meadow, that remained uncut each year until August, occurred between 5 and 35 m from the trunk and some of the mid-transect sampling points for the *Cedrus atlantica* occurred in places with heavy footfall. The sampling areas were almost flat, but with variations in elevation ranging from 30cm to 100cm and there were buildings and other tree types located in the area. The University Of Worcester has a meteorological station and a pollen trap situated within 100 metres of each tree, which allowed the prevailing wind direction to be determined for each peak pollination period.

Soil sampling

For each tree, a transect stretching up to 100m away from the trunk of the tree was sampled in the wind direction most prevalent during the peak of the flowering period. Surface soil samples were collected for distances from the base of the trunk at 1m, 5m, 10m and then every 10m after that. At each sampling point, 4cm^3 of surface material was taken from a 2m line bisecting the transect at 90^o. Soil samples comprised only the top few millimetres of the surface material. In some samples, the material was partly composed of roots, moss and leaf litter where the soil was greatly concealed by these in a matted formation. All sampling was undertaken approximately 2 months after the end of the tree's flowering period to allow sufficient time for the season's pollen to be incorporated into the surface soil/material.

Analysis of samples

From each sample, 5ml of soil was used for analysis and the pollen was extracted from the soil matrix using standard processes of hydroxide digestion, acetolysis and heavy liquid separation, as outlined in, e.g. Brown (2008) and Moore et al. (1991). The resulting pollen pellet was mixed thoroughly with 9 drops of basic fuchsin with glycerogelatine mountant (using a 1ml pasteur pipette) and spread evenly on three slides, to fit under coverslips 50 x 22 mm. On each slide, four transects, 0.5 mm wide and 48 mm long, were examined for the target pollen types. Pollen counting was done using bright-field microscopy at x400 magnification.

Modelling

The atmospheric dispersion of pollen grains from the trees used the Gaussian principle, e.g. Seinfeld and Pandis (2006). An idealised approach was followed, in a similar way to Skjøth et al. (2013), by employing the mathematical formulation from the OML model (Olesen et al. 1992) used within the DAMOS system (Geels et al. 2012). This formulation was used to simulate pollen concentrations at the surface along one transect from individual sources. A number of points located at the centre of each tree were used and from each of these a plume was emitted. Each point was placed with a vertical distance of one metre from another, from the bottom of the canopy to the top of each tree. The assumption being that each plume from the combined set of points will emit the same amount of pollen at each time step where the tree is estimated to release its pollen. The source strength of the individual points was estimated by using the shape and height of the tree, as proposed by Hidalgo et al. (1999) and later used by Aboulaïch et al. (2008). The dispersion profile for each plume therefore varied with the actual meterorological conditions such as wind speed, according to the Gaussian formulation. This resulted in each tree having a different number of plumes, as shown in Table 2. Therefore, it was assumed that the pollen production at each level is linearly related to the diameter of the tree crown at that level (Molina et al. 1996).

The height of each tree was estimated using a clinometer, as was the height of the tree at the widest point and the height above the ground of the lowest part of the canopy (Table 1). Calculations of the dispersion profile from each tree were then carried out using a subset of meteorological data from the on-site weather station with 15 minute resolution. This subset was based on the following three requirements: 1. The wind flowed along the observational transect away from the tree and must be greater than 1 m/s; 2. The time during the season should match the main pollination period as estimated from the pollen trap; 3. The time of day should correspond to the typical daily pollen emission period, which is estimated to be from 10.00 - 18.00. The outcome was a set of Gaussian dispersion curves which corresponded to the full set of meteorological conditions (wind speeds) that was present when air masses and pollen release could have caused dispersion of the pollen cloud along the observational transect. These curves were then aggregated into one curve to describe the pattern. The escape fraction of the pollen that was dispersed away from the tree was estimated by using the soil observations. The comparisons with the observations then used the calculated concentration profile at the surface, thereby assuming that deposition matches surface soil concentration and neglecting any resuspension of pollen grains from the surface, which is a reasonable assumption according to Sofiev et al. (2006). This approach is that recommended by van Leuken et al. (2016), as the more advanced CFD models require a full observational-based 3D wind field for each calculation hour.

Results

Observations

The four resulting curves from the surface soil observations for the four tree types show similarity (Fig 2), each with an initial rapid decrease in the first 5 to 10 metres, followed by a long tail of low percentage amounts, reducing to zero, or almost zero, by the 100m point. However, although the patterns are similar to one another they are not exactly the same: *Carpinus* and *Cedrus* have exponentially decreasing curves from the trunk, whereas the *Juglans* and *Platanus* curves increase to the 5m point before rapidly dropping off. For *Platanus*, the pollen was emitted more broadly away from the tree up to the 40 m point, where it then dropped to a low percentage of 3.9%. For comparison, the *Pinus* results (Fig 2) has a generally similar pattern but the influence of other *Pinus* trees in the area can be seen in the curve, notably at the 30 m and 80 m points.

Table 1 shows the total amount of pollen found in each of the four tree transects, the height and width of the trees and the percentage of pollen deposited at selected points. The majority of pollen was deposited beneath the canopy for all four trees (range: 63% – 94%) and the flattening of the curve started from around the outer edge of the canopy in most cases. The amount of pollen deposited at 50 metres was no more than 2.6% of total deposition for any tree and at 100m no more than 0.2%.

Model results

The number of plumes along the modelled transect vary between the trees from 32 (*Juglans*) to 399 (*Carpinus*), which is partly due to the length of the pollen season and the prevailing winds during the season. Total calculated deposition varied substantially between the trees as this directly reflects the prevailing winds during the length of the season. The model results all show the same pattern after about 50 metres from the tree (Fig 3). This is an exponential decreasing curve, similar to that found in the observations. *Cedrus atlantica* shows a decreasing pattern from the edge of the tree canopy while the other tree species have the peak 10-30m from the edge of the canopy. The exact shape depends on the shape of the tree crown and in particular the height of the lowest parts of the canopy. The model results also show no deposition near to the tree canopy, thereby reflecting the Gaussian principle and neglecting eventual pollen release and deposition during the filtered wind speed episodes that are below 1m/s. Pollen deposition at 50m varies from 2.09% (*Carpinus betulus*) to 0.24% (*Juglans nigra*) and at 100m from 0.29% (*Carpinus betulus*) to 0.02% (*Cedrus atlantica*). These model results are largely similar to those found in the observed results.

Discussion

Observed deposition curves

The data from the surface soil transects follow a generally similar pattern to the exponentially decreasing curves found in previous studies, as detailed in the introduction. The difference with these isolated tree sources is that the tail tends to end much sooner. Whilst it is anticipated that the general pattern of surface soil deposition would be similar for other single source tree taxa, larger source areas produce higher pollen concentrations over a greater distance, as evidenced by Bricchi et al. (2000) for *Platanus* trees. Also, within a forest, pollen dispersal is more restricted and Hicks (2001) determined that pollen deposition values can be some three times higher than those found in open forest clearings. Bunting (2005) reports that the 'relevant source area of pollen' within a closed canopy forest is in the order of 50-100m.

The observed curves for *Cedrus* and *Carpinus* began to flatten out at 5m whereas for *Juglans* and *Platanus* it was 10m. The latter two taxa also saw the curve increase between the 1m and 5m points before decreasing. These differences may be due to the fact that these trees were the tallest in the study and had the most open and widest canopies (Di-Giovanni et al. 1989). The shallower curve for *Platanus* is likely to be a factor of the height and width of the source (Kuparinen et al. 2007) and because the lawn beneath remained uncut until late in the summer, allowing more pollen to be trapped at ground level. Wind speed and persistence during pollination would also have had an effect on the patterns to some extent (Damialis et al. 2005) as an increase in wind speed directly affects the turbulence which, in turn can affect dispersal patterns (Seinfeld and Pandis 2006).

Pollen production is an important factor, varying between tree types, as well as between seasons (known as masting) (Masaka et al. 2001; Ranta et al. 2005; Jato et al. 2007). According to Molina et al. (1996), pollen production was estimated to be in the range of 188.4-302.0 x 10^8 pollen grains per metre of crown for *Platanus.* In contrast, *Juglans* had the lowest total pollen in their research at 3.4-6.5 x 10^8 (*Cedrus* and *Carpinus* were not included). Data produced by Ramezani et al. (2013) suggests that *Carpinus* is a very good disperser but an intermediate to low producer of pollen. In this study, conclusions about pollen production cannot be drawn from the total pollen count since many variables will have affected the emission and deposition of pollen into the soil.

Most of the *Juglans* pollen was deposited beneath the canopy (94%). This is known as the gravity component or trunk space component, highest where either the pollen is heavy or poorly dispersed or where the flowers died and dropped to the ground before they were able to release much of their pollen to the air (Faegri and Iversen 1992). Yiyin et al. (2015) demonstrated *Juglans* to have a relatively fast fall speed compared to other broadleaf types at 0.037 m/s. However, the results in our study may also have been a factor of in-season weather conditions during the tree's flowering period. Cold, desiccating winds and heavy rain during pollination can all have a negative impact on pollen dispersal. During the *Cedrus* flowering season in September, strong gusts of wind were repeatedly observed blowing the pollen out of the tree in smoke-like clouds but it was clear that much of it fell quickly to the ground at a distance of only a few metres. At the time of sampling, many *Cedrus* flower cones lay on the ground beneath the tree still packed with pollen. This demonstrates that only a fraction of the total pollen production is dispersed in the atmosphere, concluding that, for at least *Juglans* and *Cedrus*, the amount of pollen produced by the source tree may not necessarily equate to emission levels.

Pinus and *Cedrus* pollen grains are saccate and a study by Schwendemann et al. (2007) has demonstrated that such grains have a reduced settling speed and an increased dispersal distance compared to other pollen types. The curve for saccate grains could therefore be expected to be less pronounced. However, in this particular study, the dispersal curve from the *Cedrus* tree is similar to those with non-saccate grains.

The pollen counted in this study only represents the portion of the total emission that deposited on the ground in the locality of the tree. Pollen dispersed from a tree should largely be deposited locally although an ‘escape fraction’ (Gregory 1961) will be subject to regional and long-distance transportation under favourable circumstances, as demonstrated or modelled by various authors (Ge et al. 2004; Kuparinen et al. 2007; Izquierdo et al. 2011; Skjøth et al. 2015). Brunet et al. (2004) found pollen occurring throughout the atmospheric boundary layer during aeroplane sampling and Rousseau et al. (2006), determined that small concentrations of *Juglans, Platanus* and *Carpinus* pollen (amongst others) had travelled from North-east America to Greenland. Pollen emitted from the top of the canopy is more prone to horizontal winds and greater vertical fluctuations and therefore a greater chance of dispersing beyond the local environment (Kuparinen et al. 2007). This escape fraction cannot be detected with these observational methods. However, this limitation will not affect the overall conclusion on the exponential decreasing abundance of pollen seen in surface soil samples.

Since pollen grains are resilient to transformation due to their tough outer wall, the data almost certainly represents more than one season. Incorporation and survival of pollen grains in the soil is a dynamic process, largely involving earthworms that can move pollen both up and down within the top soil horizon (Davidson et al. 1999), thus mixing up the seasonal depositions over time.

Modelling v. observations

The modelling shows that similar pollen deposition profiles can be obtained away from the trees compared to those observed in reality. This is remarkable as the mathematical formulation of the Gaussian profile neglects both pollen deposition into the surface soil itself and variations in the surface elevation (Seinfeld and Pandis 2006). Furthermore, we have estimated the release pattern from individual trees using a regional pollen station and not made a direct detection from each of the trees in question. This means that the exact number of plumes produced for the transect in reality are not known and we have ignored micrometeorological variations due to buildings and vegetation effects. Neither have we considered the length of the flowering season specific to the individual trees, which could potentially affect the number of plumes. The standard pollen trap was used to obtain the flowering period for each of the four trees, but this detection includes the escape fraction of pollen from any trees further away (Sofiev and Bergmann 2013). All this suggests that the dominating factor for the modelled pollen profile is the exponential reduction in concentration caused by advection and atmospheric turbulence and formulated relatively simply by the Gaussian models. Secondary effects are most likely physical shape and height of the source, e.g. a cone shape in the case of Cedrus or a rounded shape in the case of the Carpinus. This shape effect is a likely cause of the increase in pollen deposition seen within the first 10-30 metres for two of the species, which was only partly replicated by the model. However, the shape effect did not seem to affect the profile further away from the trees, which has large similarities with the simulated curves.

The results match well with theoretical considerations by Sofiev et al. (2006), on the importance of turbulence, and the findings by Sommer et al. (2015) on the importance of rare pollen sources. Studies by Bricchi et al. (2000), Skjøth et al. (2013) and Hjort et al. (2016) show a tight relationship between the abundance of local sources and observed pollen concentrations. All these, as well as our results, demonstrate that a direct reproduction of calculated versus observed pollen can be realised with several types of models. However, it is of high importance to have microscale observations of both meteorology and pollen release patterns when the focus is on microscale variations, such as in forensic palynology.

Conclusions

This research demonstrates through observations and modelling that most pollen is deposited beneath the canopy for a single source tree and that, apart from the escape fraction, very little pollen is deposited more than 100m away. Surface soil pollen levels, as well as the escape fraction, can be successfully estimated using a Gaussian plume model. Tree height, width and wind speed during the pollination period were found to be the main parameters affecting deposition away from the source. This research demonstrates the strength of combining the disciplines of aerobiology (the model and the hirst-type trap) with palynology (the soil samples in this case). Further work could test the model on other tree types in different habitats and topographies.

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List of Figures and Tables

Table 2 Start and end of pollen season of selected species and the number of simulated Gaussian plumes along transect, where observed wind directions were within 45 degrees of the transect, within a 15 min time interval during daytime flowering.