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Pollen analysis of barn swallow faeces (*Hirundo rustica*)

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ABSTRACT. Pollen analysis of barn swallow faeces collected at ca. 12-day intervals since their arrival from Africa for late summer flights has been made. This study showed that through the alimentary canals of insect-consuming barn swallows, a high proportion of pollen of entomophilous plants from the close vicinity of the study area can be transmitted again into environment. Pollen of anemophilous plants, attached to the bodies of insects have also been passed to these areas. Also, grains of plants growing not only in the close vicinity but also occurring in more distant areas – several kilometres away of the study area are noted in the pollen spectra. This shows that pollen spectra from swallow faeces are representative for territories larger than it is suggested by their foraging distances.

Pollen grains passing through the intestines of barn swallows and selected insects analysed both at light (LM) and transmission (TEM) microscopy, showed in some cases ultrastructural signs of initial destruction of the endexine structure or sporadically degraded exine.

KEY WORDS: pollen, barn swallow, *Hirundo rustica*, faeces

INTRODUCTION

Transfer of pollen from flower to soil and peat/lake deposits is, obviously, of a complex nature. Pollen of wind-pollinated plants is transmitted mostly by air. The taphonomy of pollen of insect-pollinated plants seems to be more complicated. Rather exceptionally pollen of entomophilous plants may be transmitted to areas by the movement of air masses during more dynamic weather conditions (Faegri & Iversen 1975). Taphonomical studies considering only this factor showed, that pollen content of these plants is hardly perceptible or very low in the rain in the close vicinity or even at analysed sites as this is demonstrated in the case of *Hedera* and *Vitis* (Bottema 2001, Turner & Brown 2004). The main transfer agents seem to be animals – first of all pollen-consuming insects (e.g. Syrphidae) and their predators – birds, bats and other predatory insects – e.g. Odonata (Bińka 2003a). The contribution of the first group to this process is difficult to assess. It seems that it is limited to the defecations of insects on the earth surface, plants as well as on wet lake shores or marginal plants. The second intermediate link of consumers has a decidedly higher contribution in this pollen transfer, as suggested e.g. by traces of changes in pollen exines after passing through the alimentary canals of these animals (Bińka 2003a) or pollen analysis of cave deposits (Navarro et al. 2001). Some of these transfer agents in this process are probably barn swallows (*Hirundo rustica*), till quite recently very common in Europe. Today, the natural habitats of barn swallows, especially in Western Europe, have essentially become more limited.

These farmland birds can be seen most often flying around farms, where they build their nests. Usually their foraging distances are estimated to within a radius of 100 to

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500 metres (Møller 2001). If a lake basin or clay-pit with water is situated within this area, it is intensely visited by swallows. It is also possible that when searching for water or mud these birds may reach more distant basins. During the flights, barn swallows drink water and catch insects flying over or floating on the water surface (e.g. Notonectidae, Gerridae – Viettinghoff-Riesch 1955) and void excrements.

According to the analysis of feeding preferences of barn swallows (Schulze-Hagen 1970, Kożena 1979), they are interested in many groups of insects. However, these studies were focused on the food composition of the nestlings only, and not of the adults, what may have caused some simplification in the final conclusions. In the view of Schulze-Hagen, the main pollen consumers constitute about 23% of the total number of insects noted in the food samples during the breeding season.

Similar studies, according to Kożena (1979), show that Diptera are the most common group of insects consumed by swallow nestlings. Among the wide variety of families, the Muscidae (*Musca domestica*), Syrphidae and Sphaeroceridae play the greatest role. Syrphidae in this data set are represented by the most common species in Europe – *Eristalis tenax*, *Sphaerophoria menthastri*, *S. scripta*, *Episyrphus balteatus,* and *Syrphus ribesii*. The author cited tens of families found in the food samples taken during the analysis, belonging mostly to the order Diptera, as well as the Homoptera, Hymenoptera, Coleoptera and others. Some of the members of this group may transfer pollen as the first or even second agent (insect predator). It seems, however, that the main initial factors of pollen transportation are hover flies. Then, without any doubt, we can state that barn swallows consuming insects should contain pollen grains in their defecations.

The aim of this work has been to examine the potential content of pollen in barn swallows faeces. In Poland such examination is still possible, where in the eastern regions of the country a natural type of agriculture still persists, i.e. with a dominant mosaic of small fields under diverse cultivation and intervening pastures, where chemical means of plant protection are used to a lesser extent. Here, swallow nests can be found in every cow barn. The different pattern of vegetation gives a good opportunity for the population of many pollen consumers to reproduce and increase, feeding not only on wind-pollinated plants – such as *Plantago lanceolata*, and on many species of entomophilous plants. Insects consumed by predators, filled with pollen in their alimentary canals become as a consequence part of the swallow faeces.

It is also important to examine to what extent pollen exine undergoes visible transformation passing through the intestines of insects and alimentary canals of the barn swallows. Earlier, selective analyses of such faeces (Bińka 2003a) collected from nests (modern or several years old) showed the occurrence of single pollen grains of *Centaurea cyanus* with changes in the exine structure, a phenomenon often noted in Holocene deposits. Also this suggested that the removal of endexine may have taken place in the bird's alimentary canals.

MATERIAL AND METHODS

The study area (Fig. 1) penetrated by barn swallows is dominated by arable crops with rye cultivated in rotation with a mix of oat, wheat and barley, with typical weeds mostly representing Secalietea – such as *Centaurea cyanus*, *Agrostemma githago*, *Lithospermum arvense*, *Spergula arvensis*, *Rumex acetosella*, and many others, as well as fields of potatoes with weeds, dominated by members of the Chenopodietea. Smaller areas are occupied by pastures, dominated by grasses and *Trifolium repens*. However, because of intense grazing, the flowering plants in which syrphids may be interested are rarely noted, except for the massive occurrence of *Leontodon autumnalis* and *Taraxacum officinale*.

Waste lands, mostly roadsides and field margins, inhabited by many common species such as *Achillea millefolium*, *Cichorium intybus*, *Potentilla anserina*, *Polygonum aviculare*, *Artemisia vulgaris*, and members of the Apiaceae family (e.g. *Daucus carota*, *Heracleum sibiricum*) or Brassicaceae are important sites for pollen eaters. Additional sources of pollen found in the faeces are represented by flowering fruit trees, cultivated, however, on a small scale.

Faeces of *Hirundo rustica* were collected in the summer season of 2003 at Stok Ruski in the Podlasie region in eastern Poland (Fig. 1) in a small cow barn. The first comings of swallows were observed from the 1st of May. A small population of eight pairs of swallows laid two or three clutches during this period. The first traps were installed on May 6 th. The traps were made of halves of plastic pipes, about 1.5 m long, hung up by ropes under the roof in places where the swallows were observed more frequently. The traps were replaced at more or less even time intervals – 17.05, 26.05, 5.06, 17.06, 28.06, 11.07, 26.07, 10.08, 7.09 (the late summer flights of birds). 32 individual samples of the faeces were taken from each trap during time interval. After adding *Lycopodium* pellets,

Fig. 1. Map showing location of the area of investigation (rectangle) Podlasie region, Poland

the samples were treated with standard palynological procedures used in the analysis of lake/peat sediments – KOH, HF, acetolysis. When the pollen frequency was sufficient, usually one pollen slide was counted, otherwise, in the case of high pollen frequency, only its small area was examined.

The results are presented in form of a percentage pollen diagram (Figs 2, 3), with the total pollen content in each examined sample placed in the last column. For technical reasons the pollen diagram is presented in a simplified form, without excluded low-frequently noted taxa.

For transmission electron microscopy (TEM), modern pollen grains of *Centaurea cyanus* as well as these from the insect faeces were acetolysed. Grains collected from the swallow faeces were additionally boiled in 5% KOH. After that, the samples were treated with standard TEM procedures. Finally, the pollen was embedded in Spurr's low viscosity resin.

RESULTS AND DISCUSSION

Pollen from faeces noted in the slides is in a good state of preservation. Only the grains of *Centaurea cyanus* show sporadic signs of degradation of the exine structure. About 101 taxa, obviously representing a very rich set of plants are noted in the pollen spectra. About 175 samples in the pollen spectra registered a higher content of the pollen of entomophilous plants and in 100 samples the pollen frequency was low and as a rule contained rather anemophilous plants pollen. Spectra of the latter group can be recognized easily not only as samples with a low pollen content, but also as a random mix of many pollen types of entomophilous and anemophilous plants. This pollen probably was attached to the body of *Musca domestica* and consumed together with it. In the case of spectra with pollen of insect-pollinated plants, the number of grains per sample oscillated from a dozen or so to several hundred thousands.

The analysis of the percentage diagrams (Figs 2, 3) of pollen content in the barn swallow faeces indicates a large amount of anemophilous pollen, e.g. oak, pine, birch or *Secale* (partly after periods of their owering) in numerous spectra. It seems that these grains were generally not a subject of insect interest and intentional consumption, and were introduced into the swallow faeces being attached to the insect bodies.

Spectra with a higher proportion of pollen of entomophilous plants are very similar to those noted in syrphid evacuations (Bińka 2003b), however, the pollen types in this case are often more numerous (3–4 pollen taxa in the case of barn swallows against 1, 2–3 ones in the case of syrphids). This may indicate that, as a rule, fragments "produced" from two insect bodies may be found in the swallow faeces. Usually, among the recognized pollen types – similarly as in the analysed faeces of hover ies – the members of Brassicaceae and Asteraceae, especially *Anthemis* type, *Aster* t., *Helianthus* t., and in the late summer also *Artemisia* were noted. Other common components of the faeces were the pollen grains of the Apiaceae family. Its pollen curve comprised various types representing very common species growing in many habitats, e.g., *Aegopodium podagraria*, *Anthriscus sylvestris*, *Daucus carota*, *Torilis japonica*, *Petroselinum sativum*, *Peucedanum oreoselinum*, and *Cnidium dubium*. In the pollen diagram, the *Heracleum* curve was shown separately. In the spectra are also observed such pollen types as *Cicuta virosa* and *Angelica palustris*. The nearest known sites of these taxa are located about 2–3 kilometres from the study area. This might indicate distant foraging ights of the swallows or the randomly distributed migration routes of insects.

Pollen of another common family – the Rosaceae (total curve on Fig. 2) – is represented more often in the swallows faeces than in previously analysed syrphid faeces (Bińka 2003b), especially during May and June. First of all, the pollen types representing garden trees – *Prunus*, *Cerasus*, *Malus*, as well as *Sorbus* and *Potentilla* type were noted.

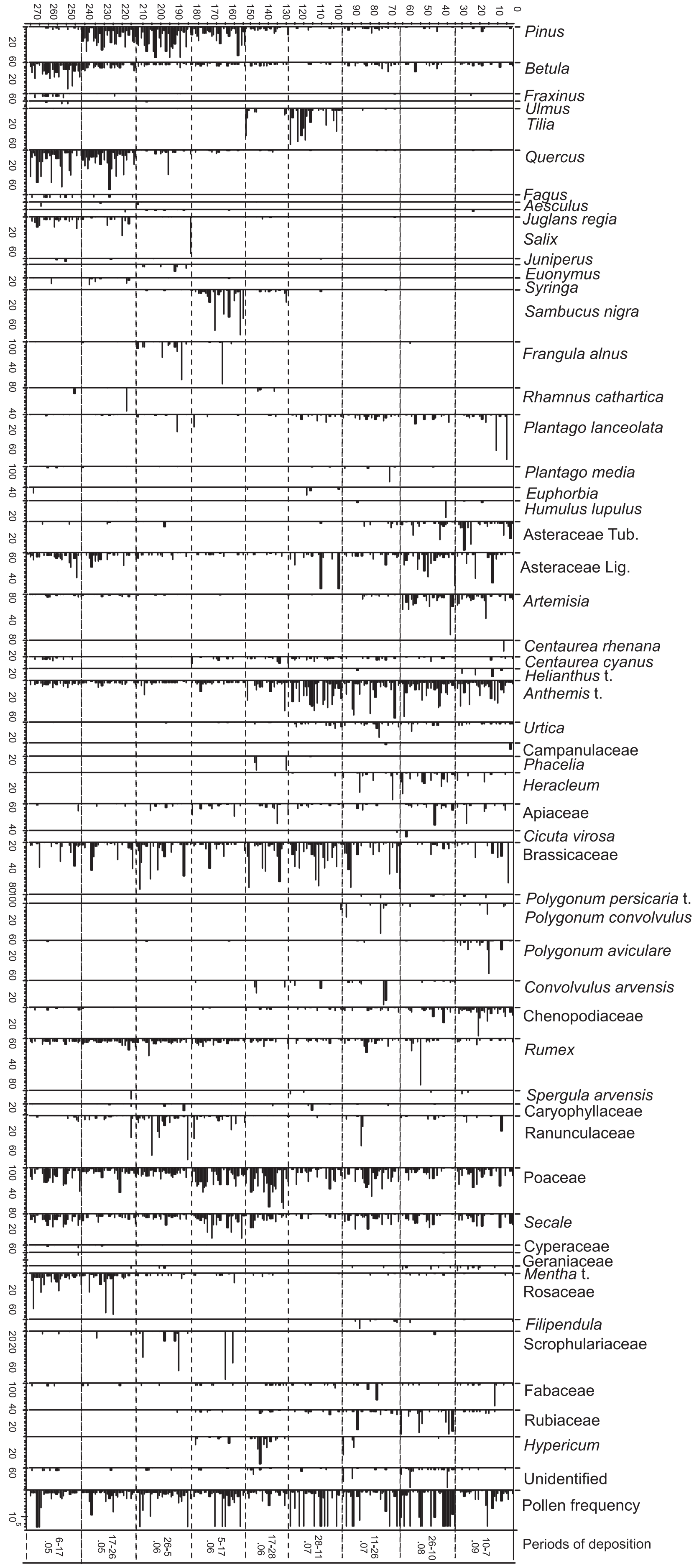

Fig. 2. Percentages of main pollen types in analysed barn swallow faeces

Pollen type more often registered in the diagram – *Tilia* and *Sambucus nigra* – seem to be over-represented in the spectra in relation to the occurrence of the plants in the vicinity of the study area. The same applies for the members of the Rhamnaceae family, *Rhamnus* and *Frangul*a, growing in the forest at the distance of 400 m from the nests.

For palynological considerations, especially those dealing with human impact on the environment registered in the Holocene diagrams, it is essential to state that apart from many pollen types applied in such inferences, numerous important indicator types occur in abundance. They are represented first of all by *Centaurea cyanus*, *Polygonum aviculare*, *P. convolvulus*, *Convolvulus arvensis*, *Spergula*-t., and many others amongst the entomophilous ones. On the other hand, more numerous are the grains of *Plantago lanceolata*, showing several culminations. This may indicate the consumption of insects, which prefer wind-pollinated plants such as *Melanostoma*, *Platycheirus*, *Pyrophaena* (Groot & Grabandt 1970). Other useful anemophilous plants, e.g., *Rumex acetosa*/*acetosella*, *Artemisia, Secale*, and Chenopodiaceae appear regularly.

The Fabaceae is a family clearly under-represented in the pollen spectra in comparison with its real population in the study area. The reason for this may be the ower structure, which is poorly accessible for common pollen consumers and low pollen production.

The pollen diagram registered quite high oscillations during the short time intervals. This is, obviously, connected with the changes of weather conditions in the study area.

In the case of barn swallows, the complex nature of using insect resources is linked to this factor. During cold weather (or increasing humidity), when the amount of insect in the open landscape is relatively low, barn swallows use mostly *Musca domestica* resources ying in barns or around cattle on the pastures.

A detailed study concerning the relationship between the climatic factors and the content of insect taxa in food samples of nestlings was made by Kożena (1979). According to this examination, increasing humidity and decreasing air temperature resulted in the higher content of Muscidae and Sphaeroceridae in the diet of nestlings. When these parameters are inverse, the syrphids in turn represent the dominant insects in the examined food samples.

PROBLEM OF PARTIAL DIGESTION OF THE POLLEN EXINES

Exines of pollen grains consumed and passed through the alimentary canals of animals undergo substantial, although in many cases difficult to observe, transformation or even partial removal. In the Holocene deposits, the pollen grains of some important species (*Centaurea cyanus*, *C. rhenana* t., *Polygonum aviculare*, Apiaceae) in part show a lack of endexine or degradation of exine, which was explained by previous pollen consumption (Bińka 2003a). Other pollen types probably also reveal such kind of transformation, however, during conventional counting the endexine is not always visible in some taxa. From a high content of this pollen in sediments we can induce that insects and their consumers can partly be responsible for transfer of entomophilous pollen into sediments (Bińka 2003a). The proportion of this atypical pollen is various in many Holocene profiles, marking surely different composition of transfer agents and undoubtedly we must to consider every time these factors during pollen analysis. Hence the need for the examination of pollen exines collected from bird faeces. Ultrastructural examination of the intestine content of selected insects (Klungness & Peng 1984, Peng et al. 1986, Suárez-Cervera et al. 1994) shows that some components of the endexine are removed in the process of digestion, although the shape of pollen and the structural elements of the exine remain unchanged. The only visible elements during the analysis of modern faeces of insects (e.g. syrphids) are rarely observed signs of "pseudogermination" marked as star-shaped structures near the endopore of the Apiaceae (Bińka 2003b). These characteristic signs we can also observe sporadically in analysed faeces of barn swallows as well as in fossil Holocene samples. Here, however, they are more frequently noted (Bińka 2003a). The reason of this may be the impairment of structural elements of endexine only in the pore areas during initial stages of germination of pollen in insect intestines than their disruption. Probably the later action of destructive factors, first of all heterotrophic bacteria in the deposits or successive consumers, causes the creation of these disrupted structures in the weakened places, open to easer penetration of destructive enzymes.

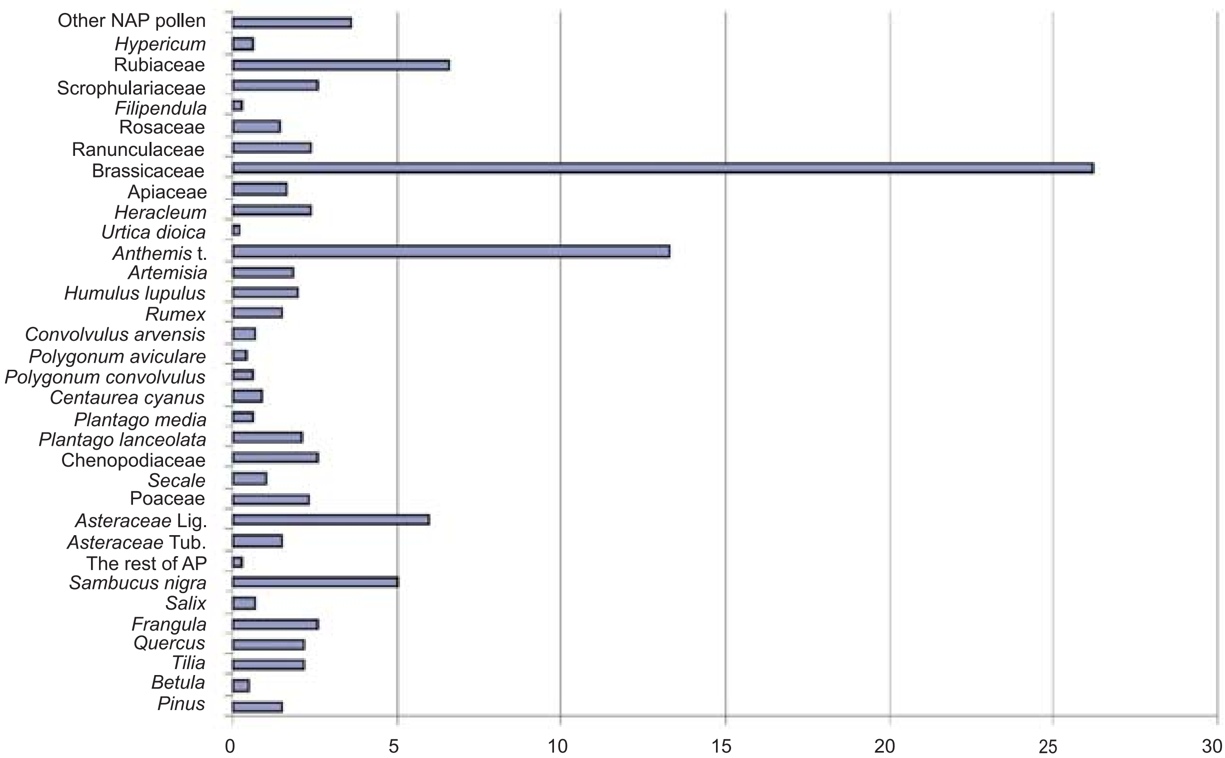

Fig. 3. Total content of main pollen types in the faeces throughout the season (percentages)

The analysis of barn swallows faeces collected in the nests (Bińka 2003a) demonstrated that some pollen grains of *Centaurea cyanus*, and *Polygonum avivulare* showed a lack of endexine. Based on this, the preliminary conclusion has been drawn that the complete destruction of endexine (its removal) or degradation of exines may take place due to the enzymatic action on pollen exine as early as when passing through the alimentary canals of the predators (e.g. barn swallows) which consume insects.

This statement has only partly found confirmation in the material studied. The pollen analysis of the collected swallow faeces does not show pollen types (especially *Centaurea cyanus* and *Polygonum aviculare*) with completely dissolved endexine. Only sporadic pollen grains of *C. cyanus* (a feature noted also in fossil deposits – Bińka 2003a) with degraded exine structure have been found. However, the endexine showed changes which were hard to identify in light microscopy. TEM analysis of modern *C. cyanus* pollen shows costae colpi build with endexine usually with small elongated empty holes parallel to foot layer in the bottom part of costae and cavities and irregularities of costae surface from the aperture side (Fig. 4: 5). Rarely these holes are larger in size (Fig. 4: 6). This construction of endexine seems to accelerate enzyme action and allow their easer penetration into the pollen.

Pollen grains of *Centaurea cyanus* from swallow faeces show in TEM analysis (Fig. 4: 1–4) reater level of destruction of costae especially in areas more vulnerable to enzyme action. This is visible as more degraded endexine (1 and 2) or lack of its fragments near the apertural zone (3 and 4).

These disintegrations seem to be not defined during TEM analysis of *C. cyanus* pollen, which passed only through the intestines of insects (*Helophilus pendulus*). This pollen does not differ structurally from modern reference material, though as mentioned above probably undergo chemical changes of endexine (Suárez-Cervera et al. 1994).

Previously collected and analysed swallow faeces (Bińka 2003a), in which *C. cyanus* pollen with dissolved endexine was noted were probably not represented by fresh material and they had being hanging attached to the nests for a few years. In humid conditions in the cow barn, heterotrophic bacteria may have caused partial destruction of the endexine. It is not excluded that the final complete destruction and removal of the endexine took place rather not in the alimentary canals of

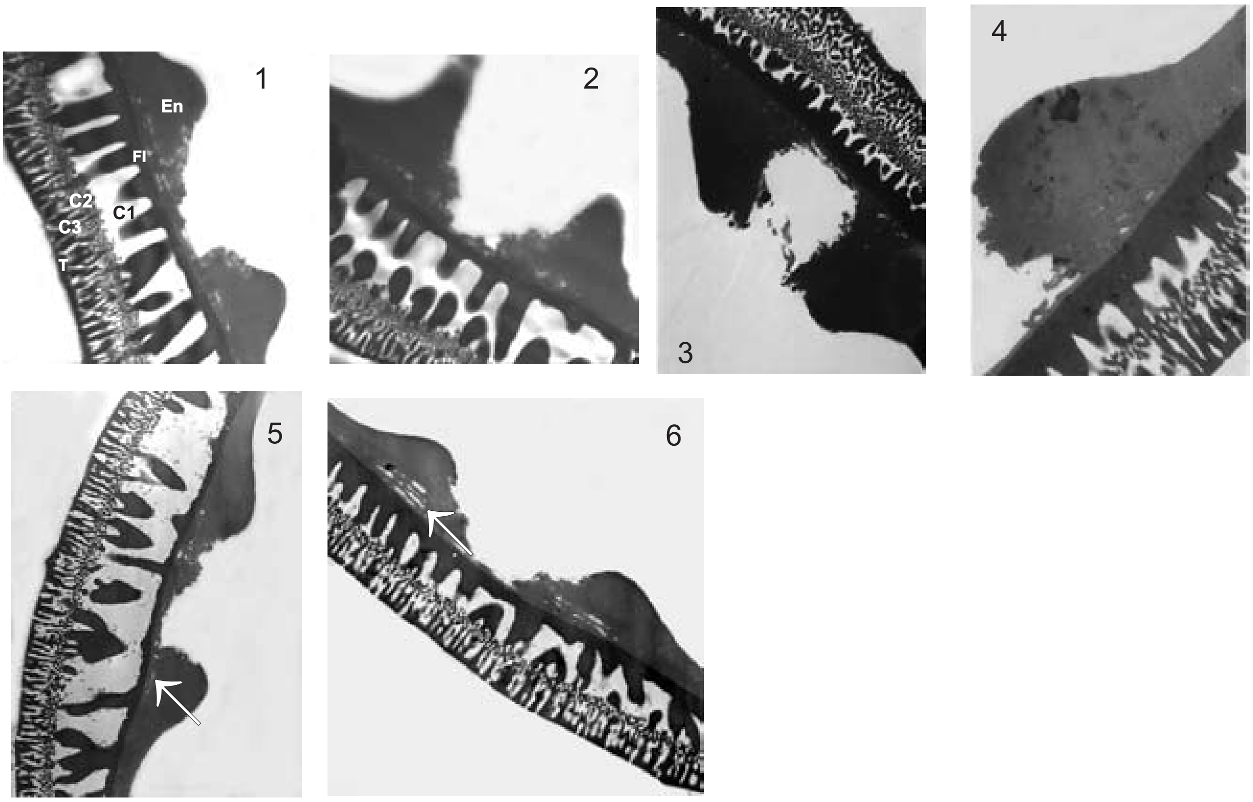

Fig. 4. 1 – 6 Pollen wall of *Centaurea cyanus* (TEM). **1–4** pollen collected from swallow faeces. Note degraded endexine near the aperture or partially removed endexine (**3–4**). **5–6** modern material of *C. cyanus* pollen. Visible empty holes near the foot layer and cavities in the surface from the aperture side (**5**). Rare type of pollen with more prominent elongated holes accelerating degradation processes (**6**). **En** – endexine, **Fl** – foot layer, **C1,C2,C3** – layers of columelle, **T** – tectum

swallows but in the areas, where faeces with pollen are deposited e.g. in soil, bottom lake deposits, due to unknown factors (a bacterial action or renewed pollen consumption e.g. by small organisms or fishes?). This must have been a quick and abrupt process, because among the microfossils sporadically only transitional forms with partial lack of the endexine are found (Bińka 2003a). The pollen analysis of unconsolidated surface deposits from the Błędowo Lake (Bińka 2003b), collected in a few points, does also not show grains "caught in act" with the proportion of typical and changed pollen, identical as in the late Holocene layers from this lake.

It is interesting that these grains, especially *Centaurea cyanus*, excellent preserved but with dissolved endexine can be found in latrine deposits (Fig. 5). Flowers of this plant (surely with pollen grains) have been intentionally consumed in the form of brew or wine, as corn ower is a medicinal plant. This means that corn ower pollen have not undergone initial exine destruction in the intestines of insects to achieve the final effect, viz endexine removal. To estimate the time interval of this destruction, human faeces filled with corn ower pollen, which passed through my alimentary canal were buried in a plastic bag in soil. After 1-year storage, about 10% pollen of corn ower had endexine dissolved by faecal bacteria during subfossilisation changes.

All this shows that the destruction and removal of endexine is possible due to different processes on land and in the water basins. The passage of pollen grains through the intestines of successive consumers – insects and birds – may significantly accelerate this.

Fig. 5. Pollen of *Centaurea cyanus*, after passing through human alimentary canal, stored for one year in soil. Note lack of endexine. **1** – colpus view, **2** – side view

CONCLUSIONS

The barn swallow, because of the scale of the insect consumption, seems to be an important factor of pollen transmission into the environment. In the pollen diagram of swallow faeces there are noted numerous pollen taxa, important in palynological considerations such as human activity.

It seems that part of pollen of entomophilous plants reach depositional basins e.g. soil, peat bogs, bottom lake sediments by means of mouths and abdomens of animals, as we can see in swallows case. Pollen endexine passing by their alimentary canals undergoes some destructional changes, however its final removal, observed often in soil samples or lake sediments, proceeds probably quickly in post-depositional processes.

An important issue is the statement that pollen grains are collected by barn swallows not only from the close vicinity of study area but also from more distant places. Because swallow foraging distances in the breeding season do not exceed 500 m – that is most flights take place within this area (Møller 2001) – responsible for this are the actively and randomly migrating insects looking for food resources. This may be demonstrated by the appearance of pollen types of such plants in the diagram, the nearest sites of which are situated about 2–3 km from the study area.

The existence of barn swallows on our territories seems to be as old as prehistoric settlement, when wooden farm buildings appeared and cattle was bred. If the second condition was not fulfilled, then the swallow population dramatically decreased (Møller 2001). Then, the lack of settlement reduces their number, although not definitely. Nests attached to various natural or artificial elements outside the settlement area can be also observed. There can be little doubt that in prehistoric times barn swallows sparsely inhabited the forest communities, surely having some influence on pollen distribution.

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