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Pollen analysis of volcanic ash in Pompeian human skeletal remains

MARTINA WEBER¹, SILVIA ULRICH ¹, ANNAMARIA CIARALLO^{2†},
MACIEJ HENNEBERG^{3,4} & RENATA J. HENNEBERG⁴

¹Department of Botany and Biodiversity Research, Division of Structural and Functional Botany, University of Vienna, Vienna, Austria, ²Archaeological Superintendency of Pompeii, Pompeii, Italy, ³Archaeology, Flinders University of South Australia, Adelaide, Australia, ⁴Adelaide Medical School, The University of Adelaide, Adelaide, Australia

Abstract

The time of the Vesuvius eruption, which perished Pompeii, Herculaneum and surrounding areas in AD 79, was initially set on the 24–25 August, based on written contemporary documents of the ancient historian Pliny the Younger. This date has been challenged by archaeologists and volcanologists/meteorologists, who moved the time of the eruption further into the autumn and eventually agreed to the final date 23–25 October. The October date has been confirmed by the latest discovery of inscriptions in freshly excavated areas of Pompeii suggesting the mid-late October eruption. In our original project of 2008 we attempted to solve the problem of the eruption time by analysing pollen mixed with falling down volcanic ash, and preserved intact in nasal cavities of the victims in Pompeii. The entire pollen spectrum, 31 different types, was evaluated with the focus on the exact time of the volcanic eruption. No date of eruption could be suggested from this study. The analysis revealed an unusually high amount of pollen from *Hedera*, an insect pollinated plant flowering from September to October in the area of Pompeii. Among three samples of ash from nasal cavities of two children and an adult considered uncontaminated *Hedera* pollen was found in noses of both children but not of the adult. This result is the first physical proof of *Hedera* as medicinal plant used for the treatment of respiratory tract disorders nearly 2000 years ago.

Keywords: *palynology, nasal cavity, Hedera, paleo-medical, Vesuvius, Pompeii*

For centuries, late August has been widely accepted as the time of the volcanic eruption that devastated the two Roman towns of Pompeii and Herculaneum and many smaller settlements at the foot of Vesuvius. This particular month was supposedly mentioned by witnesses of the tragedy (Pliny the Younger, *Epistulae*) and then accepted by many scholars, including the botanists, studying effects of this eruption (Ciarallo 2003). Earlier attempts at confirming the August date for the eruption by studying the geology and contents of alluvial deposits around the river Sarno proved inconclusive, because the deposits were mostly sterile, or even some suggested, as early as in the eighteenth century, that this eruption occurred later in the year pointing towards November as the possible date

(Borgongino & Stefani 2001). This latter suggestion of November as the month of the AD 79 eruption in which Pompeii and other settlements were destroyed, has been accepted by some archaeologists, also for the finding of a coin (Stefani 2006) of which the date of issue was later debated and considered not very legible. The discussion concerning the exact date of eruption continued and especially the August date has been challenged by volcanologists/meteorologists, who moved the time of the eruption further into the autumn when analysing the high altitude winds and the distribution of ash by these winds (Rolandi et al. 2007). After this evidence and re-examination of ancient writings most scientists and archaeologists eventually agreed to the final date of 23 to 25 October (Angela 2014). The

Correspondence: Martina Weber, Department of Botany and Biodiversity Research, Division of Structural and Functional Botany, University of Vienna, Vienna, Austria. E-mail: martina.weber@univie.ac.at

[†]Deceased

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October date has been confirmed by the latest discovery of inscriptions in freshly excavated areas of Pompeii suggesting a mid-late October eruption (communication by Soprintendenza di Pompei in BBC news, 16 October 2018). This latest discovery, although not fully published yet, seems to settle the date of the AD 79 Vesuvius eruption.

Despite the fact that with this latest discovery our main goal of using pollen for establishing the exact date of eruption in AD 79 became no longer a priority, we thought it important to continue the study of the samples from Pompeii for the purpose of testing the usefulness of specific sampling in archaeology in general.

For example, there have been problems with sampling the ash and other deposits in Pompeii for the exact dating of the AD 79 eruption. Many smaller eruptions of the Vesuvius, floods, natural movement of the ground, human activities in the area, and thus the difficulty to avoid contamination of the samples by more recent (or even earlier) materials, make the results of dating based on these samples rather uncertain.

Among possible places in Pompeii at which reliable samples of ash could be taken, are the inner spaces of human and animal skeletons. These spaces, especially if protected by a complicated anatomy, often remain unspoiled by contamination from outside materials. The second reason for that choice of sampling is, that the ash covering the body of the victim during the eruption and filling in the skeletal openings after the soft tissues decomposed, has the closest composition to the ash at the time of eruption. Some of the bodies recovered during the first systematic excavation in Pompeii, before being exposed to the outer environment were first cast by filling spaces surrounding them with plaster of Paris (De Carolis et al. 1998). Some of these casts were discarded as unsuitable for public exhibitions, but the bodies inside the casts with the ash surrounding the bones are a good source for uncontaminated samples and can be used for palynological investigations.

Nasal cavities are good sources for collecting pollen (Montali et al. 2006; Wiltshire & Black 2006). Victims immediately prior to their death inhaled the air full of ash and other materials like pollen. Some of these would be retained in nasal cavities, as demonstrated in forensic case studies (Wiltshire & Black 2006). Even in skeletonised corpses, pollen can be retrieved from the nasal cavities.

Assuming that the volcanic ash, preserved in samples collected in the earlier mentioned way, contains pollen accumulated in it during the eruption, we attempt pollen analysis in order to establish what species of plants are represented and in what quantity. This may provide us with the possible time in a year at which the eruption occurred.

We specifically addressed the following research questions: (1) Is pollen preserved intact in volcanic ash accumulated in the nasal cavities of human skeletal remains? (2) Does pollen reflect the ancient Pompeian vegetation? (3) Is it possible to define whether the volcanic event occurred in August or in October/November? (4) What else can we learn from the spectrum of pollen species recovered from individual samples?

Material and methods

Sampling

Samples of the volcanic ash for pollen analysis were chosen and collected by M. Henneberg and R.J. Henneberg from various parts of the human skeletal remains stored at the Terme del Sarno. Over approximately 250 years of excavations, more than 1000 skeletons were collected and stored in various locations. Eventually, most of skeletal materials and casts not used for public exhibitions were consolidated by the Superintendency of Pompeii in the spacious bath house of Terme del Sarno (Henneberg & Henneberg 2006, figure 1). More information about the human skeletal material excavated in Pompeii can be found in Henneberg and Henneberg (2002, 2006). The bones the samples were taken from (or around), seemed not to have been used for study before and thus had not been cleaned for the purpose of study, i.e. only partially uncovered and considered not suitable for public display casts containing skeletons which were left in storage. Eventually, samples collected from the deepest parts of the nasal cavities, after discarding the approximately 5–8 mm layer of ash blocking nasal openings, were considered uncontaminated and thus most suitable for pollen analysis, were included in this study. It seemed most likely that among any other possible places within a human skeleton the original volcanic ash inhaled during the regular breathing process at the time of eruption was still preserved trapped inside the human nasal cavity. Debris in the air inhaled through the nose or any other powder, inhaled or administered, would adhere to the walls of the nasal cavity and most likely would be there for at least several hours (Wiltshire & Black 2006).

Three volcanic ash samples for possible dating purposes are:

1. Sample 1: collected from inside a child's nasal cavity (child 1, age 2–4 years).
2. Sample 2: collected from inside another child's nasal cavity (child 2, age 2–4 years).
3. Sample 3: collected from inside an adult's nasal cavity.

4. Sample 4: surface soil sample (comparative sample). Sample 4 has been collected from the floor of the room where the human bones and discarded casts were stored. This storage room, an ancient bath, although covered with a roof had no window panes, thus it has been open to the environment. Most likely it contained subfossil pollen mixed with modern pollen and therefore represents a multi-annual deposition.

Preparation of subfossil pollen samples

Pollen was extracted and concentrated from the ash samples using the standard chemical treatments and concentration procedures for fossil pollen published by Halbritter et al. (2018). The samples were boiled in concentrated hydrochloric acid for 10 min, followed by hydrofluoric acid for 5 min, and hydrochloric acid for 5 min. The samples were then acetolysed for 5 min and treated with heavy liquid to remove the mineral

content (Halbritter et al. 2018). Glycerin was added to the remaining organic material and transferred on glass slides for light microscopic investigations. The material was observed with an Olympus BX50-F light microscope. To achieve a statistically relevant number of pollen types per sample, all taxa, excluding over-represented taxa, were counted up to 300 pollen grains. The taxon *Pinus* was overrepresented in all investigated samples. Therefore, the total amount of pollen grains varies from 428 to 3141. The results of the pollen counts are listed in Table I.

Results

The subfossil pollen grains found in the investigated volcanic ash samples are well preserved (Figure 1). A total of 31 different pollen types were found and listed alphabetically and according to their pollination type in Table I. Most pollen types are found in the nose of child 2 (27 pollen types) and the control

Table I. Pollen content of the four investigated Pompeian samples. Pollen types (plant taxa) are listed alphabetically and according to their pollination type: anemophilous (from *Alnus* to *Urticaceae*), ambophilous (from *Amaranthaceae* to *Oleaceae*), zoophilous (from *Aesculus* to *Rosaceae*). Percentage frequency of pollen types within the samples (%).

Plant taxa	Child 1		Child 2		Adult		Comparative sample	
	Count	%	Count	%	Count	%	Count	%
<i>Alnus</i>	16	1.97	13	2.25	2	0.47	62	1.97
<i>Artemisia</i>	60	7.39	26	4.49	7	1.64	87	2.77
<i>Betula</i>	7	0.86	8	1.38	5	1.17	38	1.21
<i>Carpinus</i>	0	0.00	1	0.17	0	0.00	2	0.06
<i>Corylus</i>	10	1.23	3	0.52	2	0.47	25	0.80
<i>Cupressaceae</i>	41	5.05	20	3.45	79	18.46	407	12.95
<i>Fraxinus</i>	0	0.00	0	0.00	0	0.00	2	0.06
<i>Juglans</i>	8	0.99	3	0.52	3	0.70	25	0.80
<i>Pinaceae/Pinus</i>	233	28.69	209	36.10	143	33.41	1554	49.46
<i>Plantago</i>	9	1.11	13	2.25	9	2.10	26	0.83
<i>Platanus</i>	0	0.00	8	1.38	0	0.00	28	0.89
<i>Poaceae</i>	32	3.94	18	3.11	17	3.97	85	2.71
<i>Quercus</i>	171	21.06	161	27.81	116	27.10	451	14.35
<i>Urticaceae</i>	1	0.12	1	0.17	2	0.47	3	0.10
<i>Amaranthaceae</i>	27	3.33	9	1.55	12	2.80	57	1.81
<i>Castanea</i>	5	0.62	2	0.35	4	0.93	11	0.35
<i>Mimosaceae</i>	0	0.00	0	0.00	2	0.47	2	0.06
<i>Oleaceae, inkl. Olea</i>	82	10.10	22	3.80	10	2.34	61	1.94
<i>Aesculus</i>	1	0.12	1	0.17	0	0.00	0	0.00
<i>Apiaceae</i>	7	0.86	4	0.69	1	0.23	28	0.89
<i>Asteraceae</i>	31	3.82	11	1.90	7	1.64	90	2.86
<i>Boraginaceae</i>	0	0.00	5	0.86	0	0.00	0	0.00
<i>Caryophyllaceae</i>	0	0.00	3	0.52	0	0.00	0	0.00
<i>Cistaceae</i>	0	0.00	0	0.00	0	0.00	2	0.06
<i>Dipsacaceae</i>	2	0.25	4	0.69	0	0.00	0	0.00
<i>Ericaceae</i>	2	0.25	2	0.35	4	0.93	10	0.32
<i>Fabaceae</i>	0	0.00	1	0.17	0	0.00	21	0.67
<i>Hedera</i>	57	7.02	23	3.97	1	0.23	51	1.62
<i>Myrtaceae</i>	1	0.12	7	1.21	0	0.00	4	0.13
<i>Parthenocissus</i>	1	0.12	1	0.17	0	0.00	0	0.00
<i>Rosaceae</i>	8	0.99	0	0.00	2	0.47	9	0.29
Total counts of pollen	812	100	579	100	428	100	3141	100

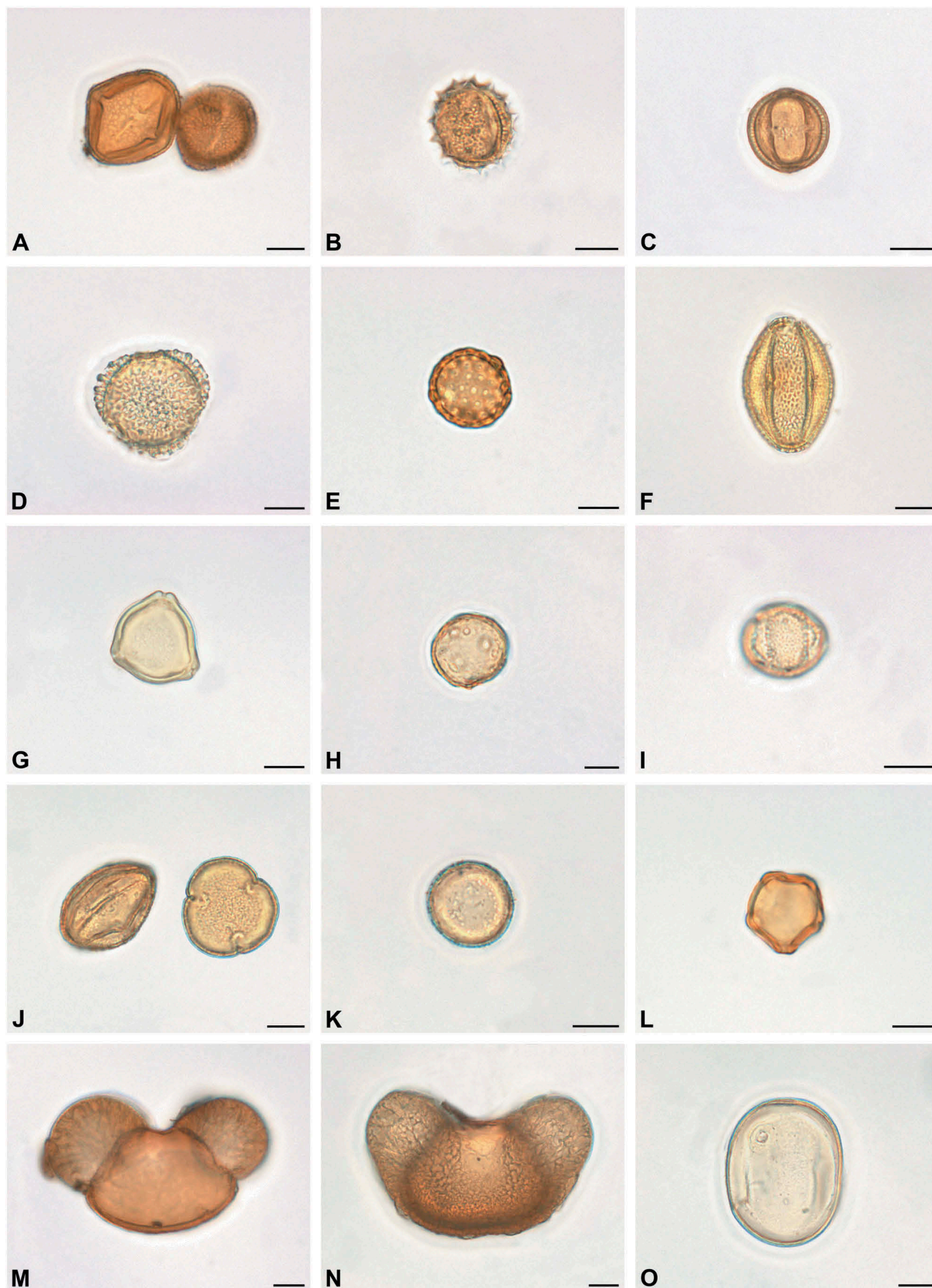


Figure 1. Most frequent pollen types preserved in the four investigated Pompeian samples. **A.** *Hedera helix*, Araliacea. **B.** Asteraceae, liguliflore. **C.** *Artemisia* sp., Asteraceae. **D.** Oleaceae. **E.** Amaranthaceae/Chenopodiaceae. **F.** *Parthenocissus*, Vitaceae. **G.** Betulaceae. **H.** *Plantago* sp., Plantaginaceae. **I.** *Platanus*, Platanaceae. **J.** *Quercus* sp., Betulaceae. **K.** Cupressaceae. **L.** *Alnus*. **M.** *Pinus* sp., Pinaceae. **N.** *Cedrus* sp., Pinaceae. **O.** Poaceae. Scale bars – 10 µm.

sample (26 pollen types), followed by child 1 (23 pollen types) and the adult sample (20 pollen types). The most common types are illustrated in Figure 1.

The pollen types of the investigated samples are very similar, but vary in quantity (Table I). The wind pollinated taxa *Cupressaceae*, *Quercus*, *Pinaceae*, and *Poaceae* are dominating, ranging up to almost 50%. The *Pinaceae* type includes the two genera *Pinus* sp. (Figure 1M) and *Cedrus* sp. (Figure 1N). Other wind pollinated taxa such as *Alnus* (Figure 1L), *Corylus*, *Artemisia* (Figure 1C) and *Plantago* (Figure 1H) occur in much smaller amounts, ranging from 1% to 7.39%. Additionally, the wind pollinated *Juglans*, is found with less than 1% in the samples. The ambophilous *Olea/Oleaceae* is found in all samples, ranging from 1.94 up to 10.10%.

Insect pollinated taxa of *Asteraceae*, *Amaranthaceae* and *Hedera*, are found in all samples, ranging from 1% to 7%. The amount of *Hedera* pollen is noticeably large in the children's nasal cavities. The insect pollinated taxa *Apiaceae*, *Fabaceae*, *Ericaceae*, *Myrthaceae* and *Rosaceae* are found with less than 1% in the investigated samples.

Pollen of the insect pollinated taxa *Aesculus*, *Boraginaceae*, *Caryophyllaceae*, *Dipsacaceae* and *Parthenocissus* is found, with less than 1%, only in the children's nasal cavities. Moreover, pollen from the insect pollinated *Fabaceae* and from the two wind pollinated taxa *Carpinus* and *Platanus* (Figure 1I) occurred with less than 1.4% only in the nose of child 2 and the comparative sample. *Fraxinus* pollen frequency is low (0.06%) and restricted to the control sample.

Discussion

Preservation of pollen

This is the first analysis of pollen preserved in volcanic ash accumulated in the nasal cavities of human skeletal remains at Pompeii AD 79. Previously, it has not been clear whether pollen grains remained intact in the pyroclastic flow that devastated the two Roman towns of Pompeii and Herculaneum. Dumbleby (2002) has reported that pollen concentrations in soil samples from the AD 79 level in Pompeii were low and pollen was poorly preserved. Gröger (2002) distinguished more than 100 pollen types, but many of them were crumpled. The first quality check of the samples in our investigation was made directly after washing the gained material from the nasal cavities. Besides primary minerals, which are typical for volcanic ash (e.g. volcanic glass), the detected pollen grains were in a good condition and coloured brown, as typical for subfossil and fossil samples. The brown colour of the pollen walls may also be a consequence of the pyroclastic flow, as heat stains

pollen walls brownish. Using the standard method for preparation of fossil pollen (Halbritter et al. 2018) we have found the pollen types preserved intact and in sufficient quantities for pollen counts. The good condition of the pollen grains in the samples from deep inside nasal cavities indicates that the bones and skin of the nose protected it sufficiently from the heat and aided the pollen preservation. Additionally, volcanic ash prevents pollen from decay, which is reported by detailed palynological studies of volcanic soils by Salomons (1986) and Fitzpatrick (1971). In such soils the active mineral agent allophane limits the biological and chemical attacks.

Pollen analysis

The catalogue of plants from the Vesuvian area lists 279 taxa, including those known from pollen studies (Jashemski & Meyer 2002). In our samples 31 pollen types were identified. All of them are in accordance with the findings in the literature (Jashemski 1979; Ciarallo & Mariotti Lippi 1993; Jashemski & Meyer 2002; Ciarallo 2004; Borgogino 2006, 2006), reflecting the vegetation as reported from ancient Pompeii. As in other palynological studies, *Pinus* is the most frequent plant in our investigated samples. The pollen composition is similar in all samples but shows significant differences in their amount.

It is assumed, that the material gained from inside the noses is a combination of pollen inhaled prior to death and volcanic ash (Luongo et al. 2003). The amount of pollen found in nasal cavities is usually small, and predominantly originating from wind pollinated plants (Montali et al. 2006). Also insect pollinated taxa can be found in nasal cavities, due to inhaling pollen within dust, soil or mud, but in much smaller quantities, as e.g. reported in forensic cases (Wiltshire & Black 2006). This is also illustrated in our samples, where diverse insect pollinated taxa are found. For example, pollen of the ambophilous olive tree is present in higher concentration compared to other insect pollinated types (Table I). This fact is noticeable, as *Olea europaea* L. is not part of the vegetation in modern Pompeii. Jashemski (1979, p. 407) also reports a larger amount of olive pollen in soil samples from 'The Garden of Hercules at Pompeii', which led to the assumption that *Olea* was cultivated in ancient Pompeii.

Most surprising is the high concentration of *Hedera* pollen in the children's nasal cavities, with 7.02% and 3.97%. On the contrary, the amount of *Hedera* pollen is small in the adult nose (0.23%) and the control sample (1.62%). The investigations by Dumbleby (2002) and Gröger (2002) showed that

Hedera pollen in soil samples covered by lapilli from Pompeii was missing or if present, the amount was small, with a mean of 0.4%. This suggests that the pollen content in the pyroclastic flow must have been low.

The discrepancy between the amount of *Hedera* pollen within the children's noses and the nose of the adult and respectively in the control sample, is strong evidence that some amount of *Hedera* pollen was already in the children's nasal cavities before inhaling ash containing pollen.

From ancient times, *Hedera* has been well known as a medicinal plant for its anti-inflammatory, antimicrobial, spasmolytic/antispasmodic properties, among other uses (Lutsenko et al. 2010). Pliny the Elder described several of its healing properties including the ability that 'took away the discomfort to the nostrils' (Ferrara et al. 2013, p. 170). Also Hippocrates and Dioscorides recommended the ivy as a cure (Berendes 1902). Young leaves as well as flowers and fruits were used to produce medicine for different purposes: bronchial asthma, diseases of nasal mucosa, bilious complaint, pertussis (in pediatrics), rheumatism, gout, icterus, ablyacousia, and even as precaution against drunkenness (Berendes 1902). Experiments in the Botanical Garden/University of Vienna have shown that pollen can end up on the leaves just below the inflorescences (Weber, unpublished data). When young leaves were used to produce medicine in ancient times, it is most likely that pollen also ended up in the medicine. In modern phytomedicine, ivy is mainly used as a medication for the treatment of bronchial diseases.

Date of eruption

Based on the pollen spectra obtained by the analysis of the volcanic ash it was not possible to define whether the Vesuvius eruption in AD 79 occurred in August or later in the year in October/November. The majority of the plant taxa found in the samples are flowering from spring to early summer. *Artemisia*, *Erica/Ericaceae* and *Hedera* are the few taxa in the sample which are flowering later in the year (Mayer 2015). The wind pollinated genus *Artemisia* is flowering from July to October/November. Due to its long flowering period, starting in July, *Artemisia* is no indicator for a late volcanic eruption. *Ericaceae* pollen was found in all samples in very low numbers (less than 1%), and based on its flowering period from March to November, this insect pollinated taxon is not relevant in this connection. The only taxon of interest in connection with the time of the volcanic eruption is the insect pollinated *Hedera*. With a flowering period from September to October,

Hedera might indicate that the eruption in fact occurred in October/November. *Hedera* pollen occurred in all samples, but in significantly larger amounts in the children's nasal cavities only. Due to the fact that the amount of *Hedera* pollen in the soil sample was much smaller with 1.62%, the high values of *Hedera* pollen in the children's noses suggest the use of *Hedera* as a medical plant in pediatrics. However, compared to other studies, the amount of *Hedera* pollen in our soil sample is also noticeably large with 1.62%. In the studies by Dimbleby (2002) and Gröger (2002) *Hedera* was present in 0.4%, and in the study by Mariotti Lippi (1995) *Hedera* occurred with only 0.2%. Given that the reference sample contains subfossil pollen mixed with modern pollen, the results cannot be used as evidence for a late volcanic eruption.

Conclusions

The analysis showed that it is possible to obtain well-preserved pollen grains, even from such extreme conditions as volcanic eruptions, when the sample is extracted from specific, anatomically protected from environmental influence parts of a human body such as a nasal cavity. This information may be useful in archaeological studies or for forensic purposes in areas of frequent geological disasters. The sampling method developed in Pompeii including the choice of nasal cavity, was used for the first time for archaeological purposes.

Two out of three samples, both from the children's noses produced *Hedera* pollen in larger quantities than the control soil sample. The presence of *Hedera* pollen in the children's noses is the first direct evidence of the treatment of respiratory conditions with an extract from *Hedera*, the practice described in ancient writings that was considered common in Roman times, also in Pompeii around the time of the AD 79 volcanic eruption.

The sample from the nose of the adult did not contain *Hedera* pollen. Thus, with our results (three samples) and no other pollen types which can be used for dating purposes we cannot establish the time of the Vesuvius eruption in AD 79 with any certainty.

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Disclosure statement

No potential conflict of interest was reported by the authors.

ORCID

Silvia Ulrich  <http://orcid.org/0000-0003-4594-3566>

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