

Recovery techniques for waterlogged archaeological sediments: a comparison of different treatment methods for samples from Neolithic lake shore settlements

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Abstract This paper presents the first comparable overview of different recovery techniques used for waterlogged Neolithic sediments in the surroundings of the Alps in the last decades. Such an investigation became necessary because it was not known which parts of plants and types of remains were absent or completely underrepresented due to inappropriate recovery techniques in Slovenian archaeobotany up to 2006. During the 2007 excavation of the approximately 5,200 years old Neolithic pile dwelling site of *Stare gmajne*, Ljubljansko barje, Slovenia, we compared three methods for the investigation of botanical macroremains: method 1 (M1) included rough wet-sieving and subsequent drying of the fractions; method 2 (M2) rough

wet sieving and keeping the fractions wet; and method 3 (M3) washing over and keeping the fractions wet. M3 with gentle washing, systematic subsampling, examination, and sorting of macroremains while wet, as well as using 0.355 mm as the smallest sieve mesh size gave the best results. When using the cruder M2 or M1 methods, waterlogged uncarbonized seeds of taxa such as *Linum usitatissimum*, *Papaver somniferum* and *Brassica rapa*, waterlogged chaff of *Cerealia* and pericarps of *Maloideae* and *Quercus* sp., which are all fragile, were underrepresented or even completely absent and therefore the plant spectra were strongly biased. On the contrary, taxa with lignified seed/fruit walls like *Cornus mas*, *Corylus avellana* or *Rubus* sp. were overrepresented when using the M2 and particularly the M1 method. The application of the M3, instead of the M1 method which has been traditionally used in Slovenian archaeobotany, helped us to identify uncarbonized remains of *Linum usitatissimum* and various species of *Triticum* for the first time in a waterlogged Neolithic site in Slovenia. Our study should contribute to a standardization of methods, which is desperately needed in archaeobotany. The study clearly shows that the plant spectra can be strongly biased if inappropriate handling techniques are used. The conclusions hold for all kinds of waterlogged sediments of different periods.

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Introduction

The accuracy of the reconstruction of past environments and people/plant relationships heavily depends on the

quality of the botanical remains and data recovered during the excavations, as well as on the quality of the analysis and interpretation of such data. The recovery of plant material on archaeological sites is influenced by specific preservation conditions (for example Willerding 1991; Van der Veen 2007), as well as by the selection of the appropriate sampling methods (for example Jacomet and Brombacher 2005 for Neolithic lakeshore settlements) and recovery techniques (for example Hosch and Zibulski 2003; Vandorpe and Jacomet 2007). It is also crucial to assess the loss due to taphonomic influence like carbonization or mineralization.

The main aim of this paper is to look at how different recovery methods for waterlogged sediments affect the representation of the types of remains and taxa and to discuss the implications of this information in terms of understanding the assemblages, as in Van der Veen (2007) which describes mainly differences between desiccated and carbonized plant remains. In this context, we will compare the method that has been traditionally applied in archaeobotany laboratories in Slovenia (Jeraj et al. 2009) and a slightly modified method with the procedure applied to waterlogged sites of the northern part of the Alpine foreland, mainly in Switzerland. The comparison of different methodologies can help to assess the quality of the results obtained as well as showing the biases that arise when sample treatment is not appropriate. An additional aim is to suggest a minimal volume of sediment samples that should be taken from the field to obtain a representative quantity of organic material in each of the fractions, such as for obtaining at least 384 identifiable plant remains for reconstructing the right proportions of the most important taxa with a probability of $95 \pm 5\%$, as proposed by Van der Veen and Fieller (1982).

The state of the art concerning the archaeobotany of Neolithic lake dwelling sites north of the Alps which existed between 4300 and 2400 cal B.C., was recently published by one of the authors (see Jacomet 2006, 2007, 2009; Jacomet and Brombacher 2005). One of the goals of this present study was therefore also to build a methodological basis for the comparison of Neolithic lake dwellings in Slovenia with those in the northern Alpine foreland.

Characteristics and formation processes of waterlogged archaeological plant assemblages, especially in Neolithic lake dwellings

Plant remains from the distant past can be preserved under water or in water-logged conditions for centuries or even for millennia. Waterlogging occurs when a material is deposited and preserved under the water table. This is the case for the prehistoric lake dwelling sites in the circumalpine region during the Neolithic (ca. 4300 to 2400 cal

B.C.) and the Bronze Age (ca. 1900 to 800 cal B.C.), among others. Much archaeobotanical research on such sites has been carried out, above all North of the Alps (for example Jacomet et al. 1989; Maier 2001, 2004; Hosch and Jacomet 2001, 2004). Such sites have shown excellent preservation conditions for waterlogged plant materials. The decay of organic plant compounds in these sites has been hindered by continuous waterlogging, low temperatures in the soil and anoxic circumstances (Retallack 1984). Therefore, most of the plant remains seem to be more or less well preserved. Such finds may include several thousand identifiable items per litre and usually 20–50 taxa per sample of <500 ml (Jacomet et al. 1989). Preservation by waterlogging, as is also the case with desiccation (Van der Veen 2007), therefore allows a much greater insight into the diversity of plant use than on average dry land sites where in most cases only carbonized plant remains are preserved. If the conditions are favourable, fragile plant tissues may survive in an excellent state, and we may even encounter green leaves when digging out freshly waterlogged sediments.

Waterlogged cultural layers contain well preserved plant assemblages which are usually mixed deposits and consist of rubbish layers mixed with dung. They represent the former “living surface” and contain two types of plant materials: they consist partly of “secondary refuse”, discarded material which had been used in some other place. Partly, however, we also observe a remarkable in situ preservation of the materials (Maier 2001; Hosch and Jacomet 2004; Jacomet 2004). Therefore, there is often evidence of discrete activities or “snapshots” of human or animal activity in waterlogged plant materials. There may be a distinct pattern in a cultural layer showing different activities (Maier 2001; Jacomet and Brombacher 2005).

Waterlogged plant assemblages include large amounts of remains connected to crop plant cleaning for the preparation of food and kitchen waste. Above all, the proportions of waterlogged cereal chaff in Neolithic lake shore settlements are high (Brombacher and Jacomet 1997). In addition, there may also be stored foods for humans and fodder for animals (Maier 2001; Hosch and Jacomet 2004), table waste and snack foods.

Besides human food there may also be remains of animal fodder, bedding, fuel, temper or building material, such as decaying wall plaster, insulation and roofing material. Such materials may include wood, mosses and also cereal straw and chaff. Animal dung and droppings as well as human faecal material, such as human coprolites from Neolithic lake shore settlements, are sources of plant material as well (Akeret et al. 1999; Kühn and Hadorn 2004; Maier 2001).

Finally, the remains of the local vegetation are present in the investigated layers as well. In the case of lake shore

settlements, these remains may have been transported by water or they were just deposited where they grew (Jacomet 1985). They can help to identify the nature of the vegetation inside the settlement or in its immediate vicinity, and may show whether a layer was deposited in the water environment or on the land. It is important to make sure that the plant remains are contemporary with the investigated settlement.

Besides waterlogged plant remains (maybe over 90%), small amounts of well preserved carbonized remains are also encountered regularly in waterlogged lakeshore settlement layers. The fewer carbonized remains may reflect what is preserved when the conditions are not favourable for waterlogged preservation. In Neolithic waterlogged settlements on average 20–50 taxa are encountered in a waterlogged state and less than ten taxa in a carbonized state (Jacomet et al. 1989). More carbonized material may be present when settlements had burnt down, like in the charred layer AH1 of the site Hornstaad Hörnle 1 at Bodensee (Lake Constance), Germany, where large cereal stores with many whole ears were preserved in situ (Maier 1996, 2001).

Materials and methods

The investigation area

All major lake and marsh prehistoric settlements in Slovenia can be found within Ljubljansko barje (the Ljubljana Moor), which is situated at the edge of the south-eastern Alps, near Ljubljana, Slovenia (Fig. 1). It

covers an area of 163 km² (Lah and Adamič 1992) and is rich in Neolithic pile dwelling settlements that existed from the first half of the fifth millennium B.C. (Čufar and Korenčič 2006) until the first half of the second millennium B.C. as revealed by dendrochronological investigations and radiocarbon dating (Velušček and Čufar 2002; Velušček 2004; Čufar and Velušček 2004). The selected site *Stare gmajne* is dated to the second half of the fourth millennium, around 3200 B.C. (Fig. 1; Velušček 2004).

The material

In 2007, a trench was excavated at *Stare gmajne*. It was 3 m wide, 5 m long and divided into 15 quadrants of 1 × 1 m². The cultural layer was 20–40 cm thick, its colour was dark brown to black with yellow clay patches, and it consisted of slightly clayey material with many waterlogged plant remains.

During the excavation, several sub-layers were distinguished. The samples for our comparative investigation come from the cultural layer between 288.87 m and 288.98 m (7th depth-cut) in Quadrants 5 and 9 (Q5, Q9). In total, six sediment samples were taken, two of 30 and four of 2 l, three from each quadrant, and each treated with a different method (see Table 1).

The 2 l samples for comparing different sieve mesh sizes were also taken from the same layer (7th depth-cut) but from the fourth quadrant (Q4; Table 2).

All methods included the same procedures of sampling in the field, subsampling in the laboratory, identification and counting.

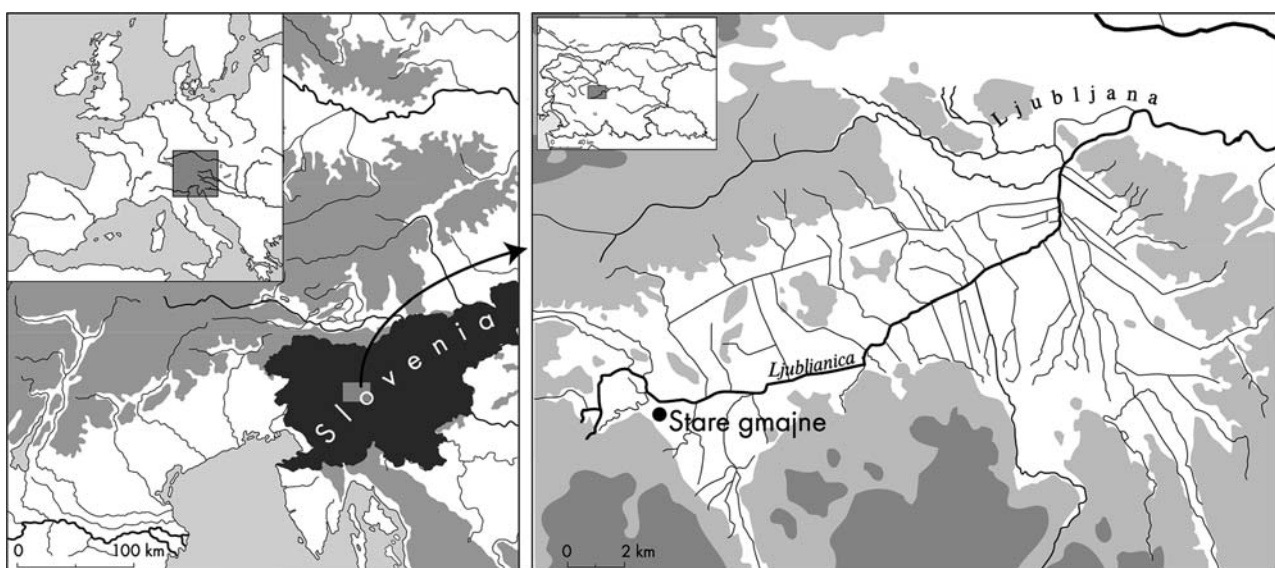


Fig. 1 Location of the Ljubljansko barje with the *Stare gmajne* Neolithic site, Slovenia

Table 1 Volumes of the sediment samples, organic fractions and examined subsamples for the compared methods: M1 dried, M2 rough wet, M3 wash-over

Method	Quadrant	Volume of the sediment samples (l)	Wet sieving method	Sieve mesh sizes (mm)	Volumes of the organic fractions (ml)		Storage	Volumes of the examined subsamples	
					Large fraction 3 mm	Small fraction 1 mm		Large fraction 3 mm (ml)	Small fraction 1 mm (ml)
M1	Q5	30	Rough	3; 1	400	150	Dry	400	25
M1	Q9	30	Rough	3; 1	500	200	Dry	450	25
M2	Q5	2	Rough	3; 1	150	70	Wet	150	25
M2	Q9	2	Rough	3; 1	180	35	Wet	180	25
M3	Q5	2	Wash-over	3; 1	280	225	Wet	280	25
M3	Q9	2	Wash-over	3; 1	530	320	Wet	260	25

Table 2 Volumes of the sediment samples, organic fractions and examined subsamples for the compared sieves with different mesh sizes: 3/1 mm (for Q5, Q9) and 2/0.355 mm (for Q4)

Method	Quadrant	Volumes of the sediment samples (l)	Wet sieving method	Sieve mesh sizes (mm)	Volumes of the organic fractions (ml)		Storage	Volumes of the examined subsamples (ml)	
					Large fraction 3 mm	Small fraction 1 mm		Large fraction 3 mm	Small fraction 1 mm
M3	Q5	2	Wash-over	3; 1	280	225	Wet	280	25
M3	Q9	2	Wash-over	3; 1	530	320	Wet	260	25
M3	Q4	2	Wash-over	2; 0.355	280	300	Wet	160	25

Method 1: rough wet sieving and subsequent drying (M1)

The first method, M1 or dry method was the one which was used in Slovenian laboratories until 2006 (Jeraj 2004; Culiberg 2006). The collected 30 l sediment samples were washed on two different sieves with 3 and 1 mm mesh sizes. The washing was done in the field as fast as possible with very rough washing and hand kneading of the clay material (Table 1; Fig. 2a).

After washing the sediment, the fractions were dried. The material was easy to store in a dry state, and the materials like bones, ceramic artefacts, charred plant remains (charcoal, cereal grains and chaff) and seeds/fruits with hard and lignified outer layers (*Rubus fruticosus*, *Corylus avellana* etc.) could be quickly picked out (Fig. 2b).

Before our analysis started, a systematic subsampling of both fractions was done (Table 1). Subsamples of 400–450 ml of the large fraction held on the 3 mm sieve and 25 ml of the small fraction on the 1 mm sieve were examined separately. These volumes gave a reliable minimal number of items in both fractions, that is, at least 384 identifiable plant remains in each fraction (Van der Veen and Fieller 1982).

Method 2: rough wet sieving and keeping the fractions wet (M2)

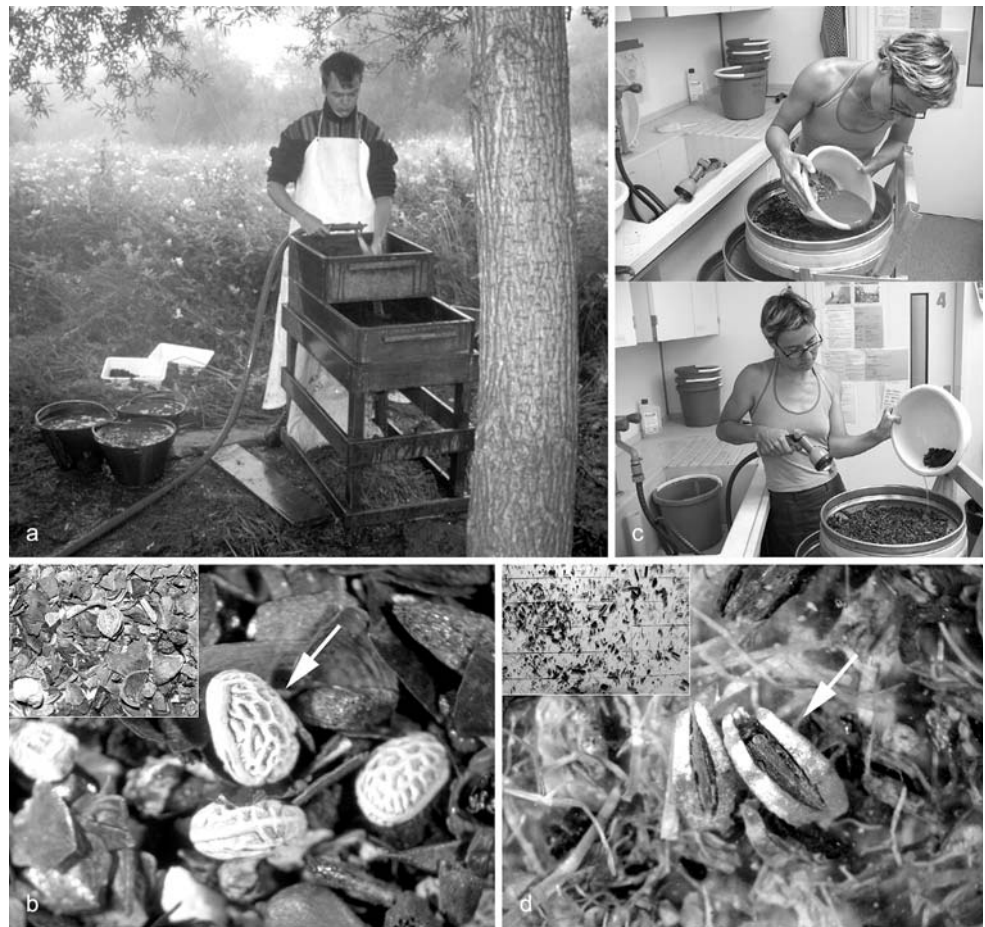
The second method, M2 or rough wet method, comprised the sieving of 2 l sediment samples in the same way as in M1 (Fig. 2a), but afterwards the collected material that remained on the 3 and 1 mm sieves was not dried (Table 1). This modification was done to find out which organic material (plant remains) was lost because of drying. The wet fractions were kept in airtight bags before the analysis was carried out. Sorting and counting were also done while the material was wet (Fig. 2d).

The subsampling was done only for the small fractions; just as in M1, 25 ml subsamples were taken and analysed (Table 1). The whole large fractions, 150 ml in Q5 and 180 ml in Q9, had to be examined, because the 2 l sediment samples taken from the field were not large enough to reach the required number of 384 identifiable plant remains (Van der Veen and Fieller 1982; Table 1).

Method 3: wash-over (flotation) and keeping the fractions wet (M3)

The third method, M3 wash-over, was described by Kenward et al. (1980) for waterlogged sediments of medieval

Fig. 2 The applied methods: **a** rough washing in the field (M1, M2); **b** dried material after sieving (M1) with hard and lignified (dried) fruit (small endocarp) of *Rubus fruticosus*; **c** gentle washing-over (M3); **d** wet material (M2, M3) with fragile and soft (waterlogged) fruit (mericarp) of *Oenanthe aquatica*



York and has been used since then in several European laboratories as at Wiesbaden by A. Kreuz and at Basel by S. Jacomet. It involves gentle washing with a sort of flotation, called wash-over (Fig. 2c). After washing the fraction of the macroremains is kept wet during all phases of work including subsampling, examination, sorting and finally storing. It was applied in the lake dwellings area in the 1990s (see mainly Hosch and Zibulski 2003). Using this method, the organic material was gently separated from the inorganic. Before sorting and counting of plant remains in water, a systematic subsampling, especially for the small fractions was necessary (see Table 1). The wet organic remains were stored and sorted in water (Table 1; Fig. 2d).

Sieve mesh sizes

In order to define which sieve mesh size is best for obtaining most of the taxa, especially for the economically important ones, the 21 sediment samples from Quadrant 4, which were also from the 7th depth-cut, were washed in the laboratory of the Institute for Prehistory and Archaeological Science (IPAS) in Basel,

where sieves with 2 and 0.355 mm mesh sizes are normally used for waterlogged sediments. For comparison, we used sieves with mesh sizes of 3 and 1 mm which were usually used in the laboratories in Ljubljana for sieving 21 sediment samples from Quadrants 5 and 9 (7th depth-cut). In both cases the M3 wash-over method was used (Table 2).

Identification and counting

Before sorting, we defined the types of the remains which were counted as one piece (“counting units”) to make sure that our target number of 384 items per fraction contained comparable remains; for justification of counting 384 remains per fraction and not per sample, see Hosch and Jacomet (2001). For cereals, we considered Hillman et al. (1996) and for other remains the system used in the Basel laboratory (Table 3; Hosch and Jacomet 2004).

When there were more than 384 plant remains in the whole examined volume, the examination of the subsample was concluded (Tables 1, 2; Van der Veen and Fieller 1982). If this was not the case as in most of the large

Table 3 Types of the remains used for identification of different taxa which were counted as 1 piece

Type of remain (“counted unit”)	Identification of taxa
Whole seed/fruit and grain	All taxa
Grain fragments with embryo end	Cerealia
Glume base	Hulled <i>Triticum</i> sp.
Rachis fragment	<i>Hordeum vulgare</i>
Fragments of seed/fruit with the tip	e.g. <i>Cladium mariscus</i>
Fragments > 1/4 of a seed	Maloideae
Fragments > 1/4 of the pericarp	<i>Quercus</i> sp., <i>Fagus sylvatica</i> , Maloideae
Fragments > 3 mm of a seed/fruit	<i>Viscum album</i>
The base of the seed/fruit	<i>Quercus</i> sp., <i>Corylus avellana</i> , <i>Malus</i> sp., <i>Trapa natans</i>
Capsule fragments > 3 mm or capsule fragments with a tip	<i>Linum usitatissimum</i>

fractions (see Table 1), the subsample had to be enlarged and sorting and counting continued until more than 384 plant remains were counted or until we ran out of the material. The latter was the case in the samples of Q5 (M1 and M2, both large fractions) and Q9 (M2, large fraction; see Table 1; ESM Table S1; Table 4).

In order to assess the comparability of the results of the M1 dried and M3 wash-over treatments, we counted all the remains, including all the vegetative parts, in two 40 ml subsamples of the 3 mm and 10 ml of the 1 mm fractions from Quadrant 9, 7th depth-cut. On the basis of the results obtained, the various percentages of remain groups were calculated (Table 5).

For examining, sorting and identifying the material, we used a Leica MZ75 stereomicroscope with magnifications of 6.3–50. For precise identification of plant remains, the reference collection of IPAS, Basel, was used and special literature (Anderberg 1994; Cappers et al. 2006; Jacomet 2008; Berggren 1969, 1981).

Table 4 Summarized archaeobotanical record per Quadrants 4, 5 and 9 (Q4, Q5, Q9) and method (dry—M1, rough wet—M2, wash over—M3): total numbers of plant groups

Quadrant	4	5	9	4	5	9	4	5	9
Depth-cut	7	7	7	7	7	7	7	7	7
Cultural layer	3	3	3	3	3	3	3	3	3
Method	M1	M1	M1	M2	M2	M2	M3	M3	M3
Sample volume in litres before sieving	~16	?	?	2	2	2	1.5	2	2
Sediment type	Clay/ org.	Clay/ org.	Clay/ org.	Clay/ org.	Clay/ org.	Clay/ org.	Clay/ org.	Clay/ org.	Clay/ org.
Large fractions									
Vol. of organic remains (ml) in the large fraction	400	400	?	110	150	180	280	280	530
Vol. of examined subsamples (ml) of the large fraction (ml)	90	400	450	110	150	180	160	280	260
Total cultivars	8	17	14	5			55	6	1
Total edible oleiferous seeds							8	4	7
Total legumes	1						2		
Total edible collected plants	20	164	288	30	44	53	206	284	351
Total plants growing outside the lakeshore		3	9				3	12	12
Total plants of the lakeshore and water plants	9	29	34	3	1	1	58	29	17
Overall Total	38	213	345	38	45	57	341	335	388
Small fractions									
Vol. of organic remains (ml) in the small fraction	?	150	?	25	70	35	300	225	320
Vol. of examined subsamples (ml) of the small fraction (ml)	25	25	25	25	25	25	25	25	25
Total cultivars	4.88	3.91	4.28	4	16	14	19	23	90
Total edible oleiferous seeds	1.49	0.37	2.05		3	4	129	28	43
Total legumes									
Total edible collected plants	13.39	63.80	18.60	67	60	209	34	53	70
Total plants growing outside the lakeshore	32.55	356.00	488.62	180	587	1,420	86	367	307
Total plants of the lakeshore and water plants	17.30	14.32	15.62	130	45	149	88	24	37
Overall total	69.61	438.41	529.17	381	711	1,796	356	495	547

Table 5 Comparison of the proportions of different remain groups, obtained by using method 1 (M1-dry) and method 3 (M3-wash-over) from Quadrant 9 (Q9), 7th depth-cut, by counting all the remains in a 40 ml subsample of the large fraction (3 mm mesh sieve) and a 10 ml subsample of the small fraction (1 mm mesh sieve), *T* thin-walled, fragile, *H* hard-walled, robust

Types of remains		M1 dry <i>n</i>	M1 dry %	M3 wash-over <i>n</i>	M3 wash-over %
40 ml 3 mm	3 mm various plant remains T	174	97.8	600	86.2
40 ml 3 mm	3 mm total seeds/fruits T	2	1.1	26	3.7
40 ml 3 mm	3 mm total seed/fruit/needle fragments T	2	1.1	70	10.1
40 ml 3 mm	3 mm overall total T without varia	178	10.7	696	67.1
40 ml 3 mm	3 mm total stones and animal remains H	584		53	
40 ml 3 mm	3 mm total different plant remains (wood, charcoal) H	760		250	
40 ml 3 mm	3 mm total other remains H	1,344	94.5	303	89.1
40 ml 3 mm	3 mm total seed/fruit/needle H	32	2.3	24	7.1
40 ml 3 mm	3 mm total seed/fruit fragments H	46	3.2	13	3.8
40 ml 3 mm	3 mm overall total H without varia	1,422	85.8	340	32.8
40 ml 3 mm	3 mm varia T & H	57		1	
40 ml 3 mm	Total seeds/fruits T & H incl. varia	34	2.1	51	4.9
40 ml 3 mm	Total seeds/fruits fragments T & H incl. varia	105	6.3	83	8.0
40 ml 3 mm	Total other remains T & H	1,518	91.6	903	87.1
40 ml 3 mm	40 ml 3 mm overall total T & H incl. varia	1,657		1,037	
10 ml 1 mm	1 mm various plant remains incl. dung T	631		3,066	
10 ml 1 mm	1 mm total Insects T	3		12	
10 ml 1 mm	1 mm total other remains T	634	94.2	3,078	91.0
10 ml 1 mm	1 mm total seed/fruit/other T without varia	38	5.6	101	3.0
10 ml 1 mm	1 mm total seed/fruit/other fragments T without varia	1	0.1	204	6.0
10 ml 1 mm	1 mm overall total T without varia	673	15.1	3,383	66.2
10 ml 1 mm	1 mm total small stones H	29		52	
10 ml 1 mm	1 mm total animal remains (bones, scales etc.) H	1,403		560	
10 ml 1 mm	1 mm total various plant remains H	1,148		931	
10 ml 1 mm	1 mm total other remains H	2,580	68.7	1,543	89.7
10 ml 1 mm	1 mm total seed/fruit/other H without varia	1,160	30.9	157	9.1
10 ml 1 mm	1 mm total seed/fruit/other fragments H without varia	13	0.3	21	1.2
10 ml 1 mm	1 mm overall total H without varia	3,753	84.3	1,721	33.7
10 ml 1 mm	1 mm varia T & H	26		8	
10 ml 1 mm	Total seeds/fruits T & H incl. varia	1,198	26.9	258	5.0
10 ml 1 mm	Total seeds/fruits fragments T & H incl. varia	40	0.9	233	4.6
10 ml 1 mm	Total other remains T & H	3,214	72.2	4,621	90.4
10 ml 1 mm	10 ml 1 mm overall total T & H incl. varia	4,452		5,112	
3 + 1 mm	Overall total incl. varia	6,109		6,149	

Grouping of the taxa

The main table presents the main results of the archaeological analysis resulting from all three processing methods (ESM Table S1). This shows the counts of all recognizable plant remains per quadrant, fraction and method (M1 dry; M2 rough wet; M3 wash-over), separately for each state of preservation (C, carbonized; N, uncarbonized; N/C, half-carbonized), including an indication of the robustness of the type of remain (for water-logged remains: T, thin (fragile); H, hard, robust; T/H, thin

to hard; C, carbonized). Other, mostly small fragments of plants like leaves, needles, roots, buds, wood fragments or mosses, etc. and parts of animals like bone fragments, fish scales, coprolites, etc. are marked as present in a semi-quantitative way (x, few; xx, small numbers; xxx, many; xxxx, very many and xxxxx, dominant).

For an easier interpretation of the results, the identified taxa have been divided into six plant groups, based on current ethnographic and ecological knowledge. This grouping is as detailed as considered necessary for our methodological approach: (1) cereals, (2) edible oleiferous

seeds (cultivars and collected wild plants like *Brassica rapa*, *Linum usitatissimum*, *Papaver somniferum*), (3) legumes (*Pisum*), (4) edible collected fruits, nuts (like *Cornus*, *Corylus*, *Malus*, *Rubus fruticosus*, *Trapa natans* etc.), (5) plants growing outside the lakeshore area (like *Abies alba*, *Chenopodium* sp., *Silene* sp. etc.), (6) plants of the lakeshore and water plants (like *Alnus* sp., Apiaceae, *Carex* sp., *Cladium mariscus*, *Mentha* sp., *Lythrum salicaria*, *Potamogeton* sp., *Oenanthe aquatica*, *Sparganium* sp. etc.; see ESM Table S1; Table 4).

Results and discussion

Comparability of the results

The comparison of the counts in the subsamples of Quadrant 9 where all the remains were counted is shown in summary in Table 5; for details see ESM Tables S2 and S3.

The material which was kept wet and treated gently with the M3 method was not richer in plant remains than the M1

dried one in terms of numbers (Table 5). However, the composition of both fractions was completely different. In M3 wash-over, fragile plant remains with thin vegetative plant tissues, leaf fragments, waterlogged cereal chaff etc. were abundant with 66–67% for all the remains in both fractions (Table 5; Fig. 3). On the contrary, in M1, the dried sample, hard remains like animal bones, wood, charcoal, *Corylus* nutshells, *Chenopodium* fruits etc. were dominant with around 85% of all the remains in both fractions (Table 5, for details see ESM Tables S2, S3). Therefore, it can be concluded that different treatments lead to different spectra of the remains so that in M1 the dried fractions, a large part of the fragile remains had changed either into unrecognizable remains or into dust and had disappeared, whereas the hard lignified remains survived and were dominant.

The results for both sieve size fractions, 3 and 1 mm, are not different in terms of the proportions of thin- and hard-walled remains: in both fractions of the M1 treatment with drying, the proportions of the robust remains are around 85%, whereas in the fractions of the M3 wash-over treatment, these proportions are around 33% (Table 5; Fig. 3). The latter figure is the real proportion of the hard-walled remains in the samples. The hard-walled remains in the dried samples are therefore overrepresented there by more than 50%; their proportion is more than two to almost three times more than their real proportion, because a large part of the fragile remains had disappeared.

But not only are the proportions of the thin- and hard-walled remains different in individual treatment groups, the spectra of taxa also differ. In the 3 mm fraction almost all the thin-walled taxa remains are found in the M3 wash-over sample. In contrast, the difference between the two treatments is not that great when looking at the hard-walled remains (ESM Table S2). This might be due to the fact that

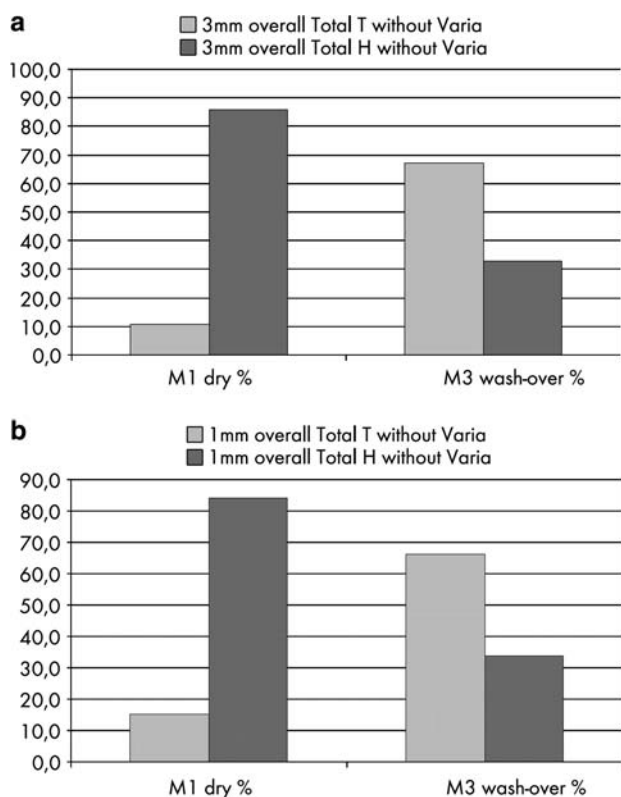


Fig. 3 Comparison of the proportions of different remain groups (*T* thin-walled, fragile, *H* hard-walled, robust), obtained by using method 1 (M1 dry) and method 3 (M3 wash-over) from Quadrant 9 (Q9), 7th depth-cut, by counting all the remains: (a) in a 40 ml subsample of the large fraction from a 3 mm mesh sieve; (b) in a 10 ml subsample of the small fraction from a 1 mm mesh sieve

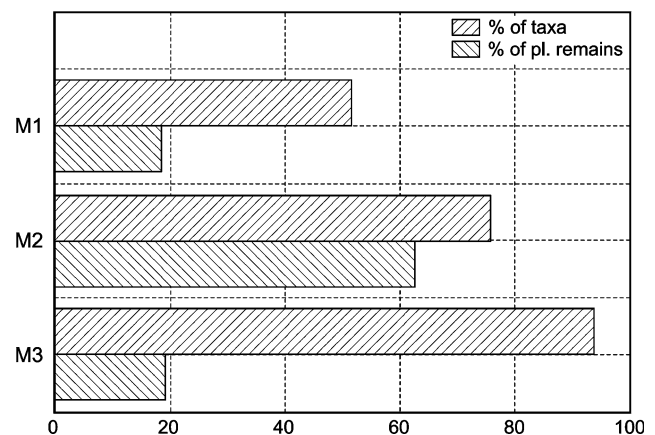


Fig. 4 Comparison of the percentages of plant remains and percentages of taxa, using method M1 dry, M2 rough wet and M3 wash-over in quadrant Q9, small fraction (1 mm)

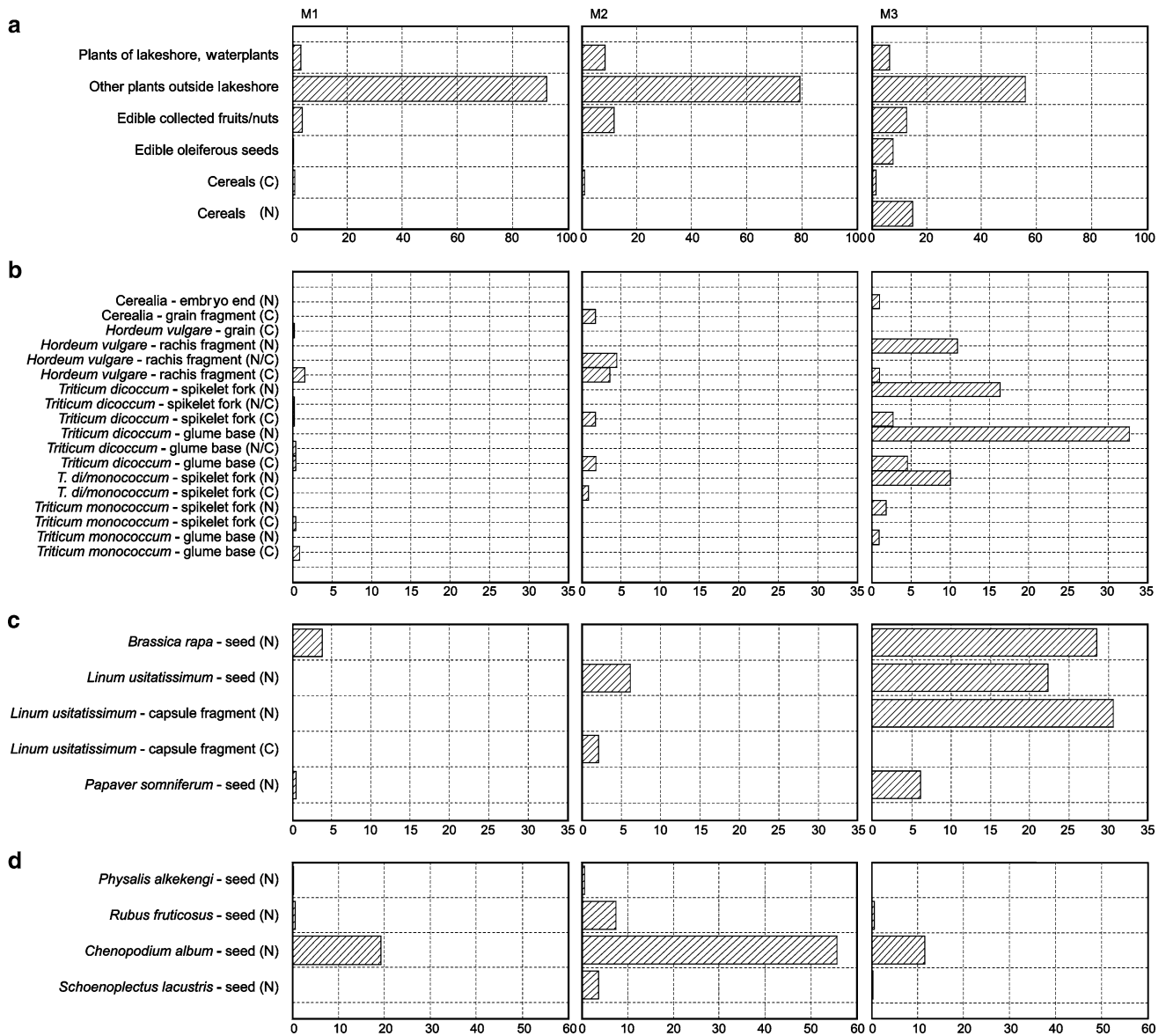


Fig. 5 Comparison of (a), plant groups and (b–d), specific taxa: (b) cereals; (c) oleiferous plants; (d) examples of plants with hard outer tissues. % of plant remains per method (M1, M2, M3) in quadrant 9, 1 mm fraction. *N* uncarbonized, *C* carbonized, *N/C* half-carbonized

in the large fraction there are more taxa with lignified seeds/fruits present like *Prunus spinosa*, *Cornus mas*, *Corylus avellana* etc.

Regarding the 1 mm fraction, the diversity of the taxa spectrum is much higher in the M3 wash-over sample (ESM Table S3). Among the thin-walled, fragile remains in the M3 sample, 20 taxa are present whereas in the M1 dried sample only six taxa were found. All the remains of waterlogged cereal chaff, flax or poppy were found in the M3 wash-over sample. Again, the spectrum of taxa is not that different regarding the hard-coated seeds/fruits. Here, we can see a distinct overrepresentation of some of the hard-coated taxa in the M1 dry sample. Extremely high numbers show a greater representation of *Chenopodium*

and also *Rubus* (ESM Table S3). This is the only case where countable seeds/fruits reach a proportion of 26.9% in contrast to 5% in the M3 fraction (see Table 5; ESM Table S3). Therefore, in order to obtain comparable values for the different treatment methods M1 and M3, the counts of the dried small M1 fractions had to be multiplied by 0.186 (obtained by dividing the proportion values of the M3 sample with those of M1 ($5 \div 26.9 = 0.1858736$)).

The results of the archaeobotanical record according to the various quadrants and methods are presented in Table S1 (ESM). The comparable value column presents the multiplied and therefore comparable values for the M1 dry samples for the small fractions. In the following, we rely on these values (Figs. 4, 5).

Representativity of the results

Large fractions

The volumes of the examined (sub)samples processed with each method differ in the large fractions (3 mm) of Quadrants 5 and 9 (Tables 1, 4). Not all the sediment samples taken in the field were large enough to reach the required number of 384 identifiable plant remains (Van der Veen and Fieller 1982). However, it becomes clear that the number of identifiable plant remains depends mostly on the treatment method. With the M3 wash-over method the required numbers were almost reached even with comparably small sediment samples of 1,500–2,000 ml. This was not at all the case when the samples were treated inappropriately. The M2 rough wet method turned out to be especially the worst—the numbers reached were less than 57 items per large fraction (ESM Table S1; Table 4). The numbers reached with the M1 drying method seemed to be better at first glance, but the sample volumes were much higher (over 10 l). Unfortunately, these volumes were not processed in two cases, Q5 and Q9, where the numbers of remains were the highest. To summarize, only the large fractions of Q9 and treatments M3 and M1 are to some extent comparable in terms of their taxa spectra because they contained >345 identified plant remains.

The volumes examined of the M1 dried (sub)samples were large, 400 ml in Q5 and 450 ml in Q9 (see Tables 1, 4). Despite this, the numbers of taxa (18 in Q5 and 16 in Q9) and identifiable plant remains (213 in Q5 and 345 in Q9) were lower than in the M3 wash-over subsamples (31 taxa and 335 plant remains in the 280 ml of examined subsample from Q5 to 19 taxa and 388 plant remains in the 260 ml of examined subsample from Q9) (see ESM Table S1, Table 4). Therefore, in cases of inappropriate treatment much more material has to be checked to provide a large enough amount of remains, in order to obtain a reliable number of taxa of seeds/fruits. All in all, the M3 wash-over (sub)samples were by far the best for obtaining a representative number of remains and taxa in the large fraction.

Small fractions

The volumes of small fractions from the 1 mm sieve from each of the M1, M2 and M3 treatments in all three quadrants were large enough to provide subsamples in which the required number of remains (>384) could be attained (ESM Table S1, Table 4). Even when the numbers counted in the M1 dried subsamples were divided by 0.185 (see Table 4) they were high enough with one exception (Q4; Table 4). Therefore, these results are mostly representative and comparable. The volume of the examined organic

material in subsamples with each method was 25 ml in all three cases (see Tables 1, 4). In the following, therefore, only the results of the small fractions (Figs. 4, 5) are considered.

Results for the small fractions of the samples from Quadrants 5 and 9

In order to highlight the most important results we have chosen the Quadrants 5 and 9 which gave very similar results (see ESM Table S1). The results are therefore presented together, and some examples of the results for Q9 are highlighted (Figs. 4, 5).

Figure 4 shows that the M3 wash-over sample in Q9 contained by far the largest proportion (94%) of recognizable taxa, although only relatively few remains (under 20%) were counted. The opposite result is shown by the M2 rough wet sample in Q9 where over 60% of the remains had to be counted in order to obtain a higher amount of taxa.

ESM Table S1 and Fig. 5a show that the M1 dry and the M2 rough wet samples have a similar pattern: most of the remains consist of the group “plants outside the lakeshore”, which are mostly weeds, above all from the Chenopodiaceae, with hard-coated seeds/fruits in proportions, based on numbers of remains of 80–90%. Moreover, edible collected fruits and nuts and plants of the lakeshore are relatively well represented, with a maximum of 10%. Seeds of edible oleiferous plants and waterlogged uncarbonized cereal remains are absent; carbonized cereal remains are represented in very low amounts.

In contrast, the M3 wash-over sample shows much more balanced picture: the group “plants outside the lakeshore” is still the most common, with slightly more than 50%. However, the other groups are represented much more evenly. It can therefore be stated that the M3 method is the only one which gives a balanced spectrum of the taxa.

If we look more closely at the taxa spectrum, the differences between the methods and their influence on the spectra of taxa represented become even clearer. On the basis of other work on material from Neolithic lake shore settlements (Hosch and Zibulski 2003), it is known that remains and taxa with very delicate tissues like waterlogged cereal chaff are destroyed and disappear when samples are not appropriately treated (Fig. 6a). This is well visible in Fig. 5b: waterlogged uncarbonized cereal remains are only present in the M3 sample; in addition, they represent the largest part of all the cereal remains. Carbonized cereal remains, in contrast, are present in all three treatments of Q9, always in low amounts. The spectrum diversity of the cereals does not differ between individual treatments; all of them contain *Hordeum vulgare*, *Triticum dicoccum* and *T. monococcum*. If we

Fig. 6 **a** Thin and fragile non-carbonized remains: cereal remains: *a* *Triticum dicoccum* glume base and *b* spikelet fork; *c* *Hordeum vulgare* rachis fragment. Seeds and fruits of oleiferous plants: *d* *Linum usitatissimum* seed; *e* *Brassica rapa*; *f* *Papaver somniferum*; *g* *Linum usitatissimum* capsule fragment, *h* *Malus* sp. pericarp fragment; *i* *Quercus* sp. pericarp fragment. **b** Hard and lignified seeds/fruits: *a* *Cornus mas*; *b* *Corylus avellana*; *c* *Crataegus monogyna*; *d* *Prunus spinosa*; *e* *Vitis vinifera* ssp. *sylvestris*; *f* *Quercus* sp.; *g* *Fragaria vesca*; *h* *Rubus fruticosus* agg.; *i* *Physalis alkekengi*

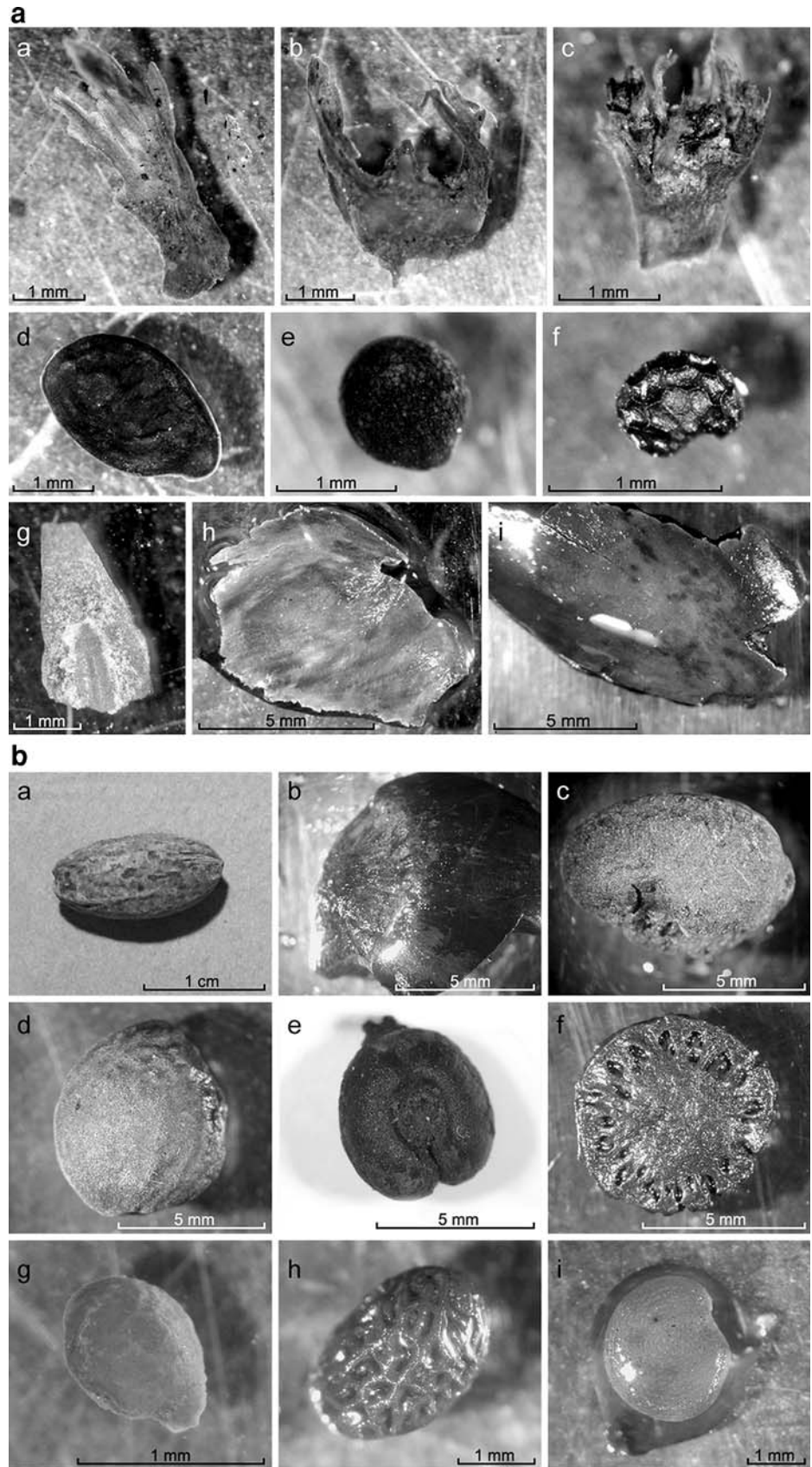


Table 6 Comparison of plant remains obtained from different sieves, small wash over fractions (0.355 mm-Q4 and 1 mm-Q5, Q9): plant group, volume of the examined subsample, total number of plant remains (Q4, Q5, Q9), number of taxa, preservation (Pres.): C-carbonized, N/C-half carbonized, NC-noncarbonized (waterlogged), seed/fruit cover (Sc) (hardiness of the outer tissue of the diaspore), *T* thin, *H* hard, *T/H* thin to hard

Group of plants	Q4—0.355 mm	No. of taxa	Q5—1 mm	No. of taxa	Q9—1 mm	No. of taxa	Pres.	Sc
Volume of the subsample	25 ml		25 ml		25 ml			
Total no. of plant remains	356	28	495	18	547	31		
Cereals	19	2	23	3	90	3		
	12		16		81		NC	T
	7						N/C	C
			7		9		C	C
Edible oleiferous seeds	129	2	28	3	43	3		
	95		17		29		NC	T
	35		11		14		NC	T/H
Edible collected fruits/nuts	34	6	53	5	70	9		
	26		11		35		NC	T/H
	8		42		35		NC	H
Other plants outside the lakeshore	86	8	367	3	307	6		
	2						NC	T
	1		5		6		NC	T/H
	83		362		301		NC	H
Plants of the lakeshore, water plants	88	10	24	4	37	10		
	55				2		NC	T
	15		14		27		NC	T/H
	18		10		8		NC	H

look at their proportions based only on the carbonized remains, the differences do not seem to be very important. There are, however, differences if we include uncarbonized chaff in the calculations: by far the most important cereal—at least in Quadrant 9—was *Triticum dicoccum* (emmer). This clearly shows that without considering uncarbonized waterlogged cereal remains, our conclusions regarding the importance of cereals would have been strongly biased. Only M3 wash-over treatment provides a reliable picture of cereals.

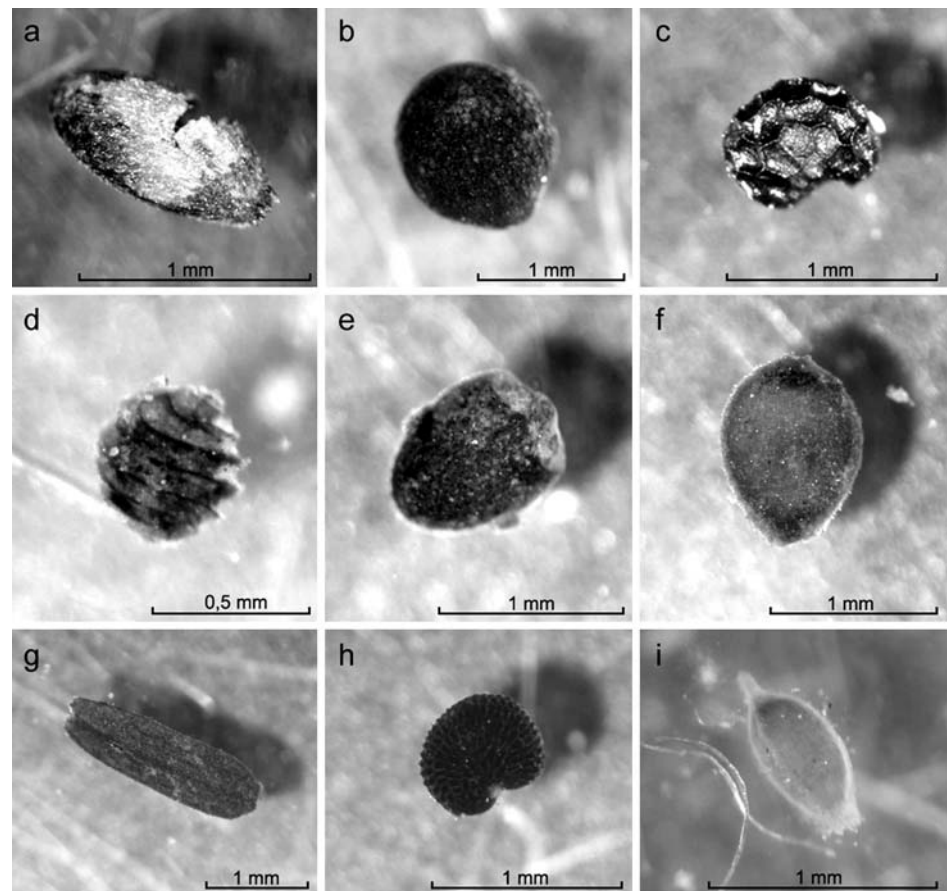
A very similar statement can be made regarding other seeds/fruits with fragile coats, especially the remains of oleiferous plants like *Linum usitatissimum*, *Brassica rapa* and *Papaver somniferum* (Figs. 5c, 6a). Most of them—or even all of them in the case of poppy seeds—are only present in the M3 wash-over sample. Their presence in the M1 dry and M2 rough wet samples is only occasional. Quite the opposite can be found out if we look at the proportions of the taxa with hard-coated lignified seeds/fruits which tend to be overrepresented in the M1 treated samples (see above, under “comparability of the results”). They survived drying and rough washing well (Fig. 2a, b) and became overrepresented because they were almost the only “survivors” when samples were badly treated. The

degree of overrepresentation is shown by the proportions of *Physalis alkekengi*, *Rubus fruticosus*, *Chenopodium album* and *Schoenoplectus lacustris* in Fig. 5d (pictures, see Fig. 6b). Even more stable carbonized cereal remains can show a certain degree of overrepresentation. This is visible in Q5 where they were present in larger amounts in the M1 and M2 treated samples (see ESM Table S1).

Differences between the sieve mesh sizes

There were no obvious differences between the results obtained when using a 2 mm mesh sieve used in IPAS Basel University and the 3 mm mesh sieve as used in Slovenia, for the large fractions (see ESM Table S1). Nothing was lost if 3 and 1 mm sieves were used instead of a 2 mm one. However, there may be differences between the results from using a 0.355 mm sieve as at IPAS Basel University, and a 1 mm sieve as was used in Slovenia before introducing new methods. It is of some importance to know what exactly has been missing from the Slovenian spectra until now, in order to compare earlier results with the modern ones. Therefore, we conducted a small, additional study to find out the differences. For this purpose, the 0.355 mm fine sieved fraction

Fig. 7 Small waterlogged seeds: **a** *Lythrum salicaria*; **b** *Brassica rapa*; **c** *Papaver somniferum*; **d** algae oospores *Chara* sp., *Nitella* sp.; **e** *Mentha arvensis/aquatica*; **f** *Urtica dioica*; **g** *Eupatorium cannabinum*; **h** *Arenaria serpyllifolia*; **i** *Cyperus fuscus*



of Q4 and the 1 mm fractions of Q5 and Q9 were compared, both treated with the M3 wash-over method (see Tables 2, 6; for detailed identifications of plant remains see ESM Table S1).

Table 6 shows that the sample from Q4, using the 0.355 mm sieve, contained many more small-seeded remains than expected. In the group of oleiferous plant remains especially *Papaver* and Brassicaceae were much more frequent in Q4 than in Q5 and Q9 where the 1 mm sieve was used. There was also an obvious difference in the number of remains of small-seeded plants, growing on the lakeshore or in water, for example *Lythrum salicaria*, *Mentha arvensis/aquatica*, *Urtica dioica* or the oospores of *Chara* sp. and *Nitella* sp., where once again Q4 using a 0.355 mm sieve contained many more such plant remains (for examples, see Fig. 7). The results show that taxa with small seeds, also including *Eupatorium cannabinum*, *Verbascum* sp./*Scrophularia* sp., *Camelina microcarpa* and *Arenaria serpyllifolia*, are generally not properly represented or even absent when only using 1 mm as the finest meshed sieve, as in Q5 and Q9 (for more details, see also ESM Table S1). Therefore spectra from Slovenian settlements, where 1 mm sieves were the finest meshes used, are

not comparable with modern studies in terms of the recovery of small seeded taxa.

Conclusions

The comparison of the M1 rough wet sieving, M2 wet sieving, and M3 wash-over methods for preparation and subsequent treatments, M1 with drying, versus M2 and M3 kept wet, of macrobotanical remains from waterlogged archaeological layers clearly confirms that the M3 wash-over method is the best one. When using the gentle wash-over procedure, many vegetative plant remains such as mosses, leaf and needle fragments, stalks, various epidermis etc. were very well preserved. In addition, fragile reproductive plant remains, above all waterlogged cereal chaff but also pericarps as well as seeds/fruits of oleiferous plants were also present in large amounts. Fragile animal remains like insects, molluscs, coprolites, fish scales etc. were also very well represented. All in all, the results of Hosch and Zibulski (2003) from Swiss Neolithic lake shore settlements could be corroborated. In addition, it could be shown that the finest sieve mesh which should be used is

0.35 mm, for obtaining a proper representation of small-seeded taxa, at least those which are of economic importance; for other problems it might be appropriate to use even finer sieve meshes. For future studies, not only of Neolithic waterlogged materials but in general, we would strongly recommend the use of the wash-over method, already described in 1980 by Kenward et al. which, however, is not applied regularly and everywhere. If the sediments are strongly compacted, we recommend freezing and thawing as a gentle and easy pre-treatment method (Vandorpe and Jacomet 2007).

In contrast, in the M1 and M2 treated samples the fragile remains were missing to a large extent, and hard, lignified and partly also charred seeds/fruits were concentrated and overrepresented because their rigid outer layers protect them from shrinking during drying and damage during rough washing, both of which damage the material. A comparison of the numbers of all the counted remains in two subsamples allowed us to estimate the degree of overrepresentation of hard, lignified remains in the M1 treated samples in comparison with the gentle M3 treatment: it is over 50%.

There were also clear differences between the methods concerning the diversity of taxa obtained. By applying the gentle M3 method and using 0.355 mm as the finest sieve mesh size, the diversity of taxa was by far the highest and only with this method was it possible to establish reliable proportions of useful plants. Using the gentle M3 wash-over method it was possible to find uncarbonized waterlogged remains of *Linum usitatissimum* (flax, seeds as well as capsule fragments) and chaff of *Triticum* species for the first time in a Slovenian Neolithic site.

As far as sample volumes are concerned, we can recommend the use of at least 3 l sediment samples for Neolithic lake shore settlements when preservation is good. This volume and the use of the M3 wash-over method make it possible to reach the required number of identifiable plant remains in both the 3 mm and the 0.355 mm fractions for a representative result. A large volume is especially needed in order to gather enough identifiable and countable remains in the large fraction. For the small fraction, the analysis of a small subsample of about 25 ml may already provide enough identifiable material. This result corroborates the conclusions by Hosch and Jacomet (2001) based on the material from northern Alpine Neolithic lake dwellings.

Our study should be seen as a contribution to a desperately required standardization of methods in archaeobotany. It clearly shows how the use of inappropriate methods can severely bias the plant spectra obtained. Moreover, it shows that a clear description of the methods applied in archaeobotanical studies is very important. With our study we are now able to judge what might be missing

from earlier archaeobotanical studies carried out in Slovenia. By using the M3 wash-over method it is possible to produce reliable data which are comparable with those from the northern Alpine Neolithic lake shore settlements for the first time.

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