1 The bigger the better? On sample volume and the representativeness of

2 archaeobotanical data in waterlogged deposits

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4 Crossreferences

5 Abstract

6 This paper provides a reference to estimate the representation of large-sized items (seeds 7 and fruits, mainly) in samples of larger and smaller volume in wetland sites with the aim of 8 proposing a minimum sample size to recover these remains in a representative way. For this, 9 almost 100 samples from a late Neolithic settlement phase found at the lakeshore site of 10 Parkhaus Opéra (Zürich, Switzerland) were subsampled into one larger subsample (A-11 sample, of ca. 3 litres of volume) and one smaller subsample (B-sample, of ca. 0.3 l of 12 volume). We compared how large and small-sized items were represented in the different 13 fractions of large and small subsamples on the basis of ubiquity, concentration and 14 proportions between the taxa. Large-sized remains (like Prunus spinosa or charred 15 fragments of Corylus avellana) and some medium-sized remains (Najas, Aethusa cynapium) 16 were more often represented in larger subsamples and therefore are considered to be 17 underrepresented in smaller samples. Average concentration values were similar in both 18 groups of samples (and therefore comparable) but large differences were observed on a 19 one-to-one sample basis, finding no positive monotonic correlation between them. Our 20 observations also prove that in order to obtain data that are comparable to dryland sites 21 concerning charred remains (including cereals and large-seeded wild fruits), large volume 22 samples of at least ca. 3 I are needed. Counting units per taxon in each fraction were re-23 defined on the basis of the results obtained. Finally, some clues to interpret results 24 concerning large-sized items in sites with samples of small volume are also proposed 25 following our observations.

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27 Key-words: lakeshore settlement, methodology, sample size, quantification

28

29 **1. Introduction**

30 Sampling in archaeobotany is a major issue, playing a key role in the interpretation of 31 botanical assemblages. When designing a sampling strategy, one needs to take into 32 consideration which contexts are sampled, their potential richness in botanical 33 macroremains, the size of the samples and the number of contexts sampled per site, in order 34 to have a dataset that can be considered representative of the total amount of botanical 35 macroremains preserved until today. Above all, the scientific questions that are aimed for 36 should be clearly stated beforehand in order to plan the sampling strategy accordingly (see 37 overviews on this issue in e.g. d'Alpoim Guedes and Spengler, 2014; Filipović and Marić, 38 2013; Jones, 1991; Lennstrom and Hastorf, 1992; Pearsall, 2015; van der Veen, 1985). 39 Archaeobotanical research in waterlogged deposits of prehistoric lakeshore settlements has 40 some specificities. To start with, sampling is usually performed before any archaeological 41 structure is identified with certainty, since this is mostly done at a second stage, after the 42 conduction of dendrochronological analyses. This means that systematic or random 43 sampling (see e.g. Hosch and Jacomet, 2001) is absolutely necessary to have different

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structures properly represented in the samples. Secondly, sample size is another important
issue. When preservation conditions are good, plant macroremains appear in extremely high
numbers (thousands in each sample). For this reason, a balance needs to be found between
having samples large enough to have all kinds of fruits well-represented in them, and at the
same time trying to analyse them in the most efficient way possible (Jacomet and

49 Brombacher, 2005; Kenward and Hall, 1995: 454-455; Steiner et al. in press).

50 Most of the research in (mostly Neolithic) lakeshore settlements done in the seventies and 51 the eighties of the XXth century was based on profile (monolith) samples (e.g. Jacomet, 52 1980; Jacomet et al., 1989; Maier, 1988; Schlichtherle, 1985), although there were some early exceptions of surface sampling (Jacomet, 1981). Profile sampling yielded samples of a 53 54 relatively small volume (mostly below 0.3 l) and recommendations were done to take, in 55 parallel, a certain amount of bulk samples (10-20 samples of more than 0.7 I per settlement phase) in order to record the large-sized items in a representative way (Jacomet et al., 1989: 56 57 82). The large research project carried out at Arbon Bleiche 3 in the early nineties made it 58 possible to recover samples of a larger volume to test if large-sized items (those taxa with 59 seeds of well above 2 mm in size or other items like spikelets or capsules) were better 60 represented in them. It was soon observed that samples of ca. 0.3 I only allow a 61 representative evaluation of small-sized items (below 2 mm) and that samples of at least 2 l 62 were recommended for a fully representative analysis (Brombacher and Jacomet, 1997: 222). The goal was to reach a statistically representative amount of remains for a sample 63 (sensu Van der Veen and Fieller, 1982). It was observed that ca. 400 remains per fraction 64 (2mm and 0.35mm) were needed for a representative analysis of a sample, so that large-65 66 sized items were also representatively recorded (Hosch and Jacomet, 2001). This made it 67 clear that larger samples were needed to reach this amount of large-sized remains in the 68 2mm fraction. As methodological conclusions of the Arbon Bleiche 3 project, it was 69 recommended (parallel to profile sampling, which remains as the optimal strategy to target 70 layer formation processes in lakeshore settlements) to take large-volume samples (ca. 3 l, 71 and a maximum of ca. 8 l) in a systematic way over the excavated surface of the settlement. 72 From these large samples, small-volume subsamples (ca. 0.3 l) could be produced in a way 73 that large samples only needed to be investigated for large-sized items (and therefore sieved 74 with a mesh of 2mm) and smaller samples for small-sized items (sieved with a mesh of 0.35mm) (Hosch and Jacomet, 2001; 2004: 116). This time-saving strategy was finally 75 applied to the recently excavated multi-phase site of Zürich-Parkhaus Opéra, our case study 76 77 (Antolín et al., 2015; 2016; 2017; Bleicher and Harb, 2015) and recently also critically revised 78 (Steiner et al., in presss).

79 In parallel to this line of research developed at the IPAS (Integrative Prehistory and 80 Archaeological Science, University of Basel), other researchers developed alternative 81 sampling strategies, like systematic coring (every meter). This type of sampling was usually 82 performed within scientific research projects (not rescue excavations) and resulted in a large amount of samples of less than 0.3 l of volume in average, or occasionally more, like at 83 Sipplingen (0.7 I in average) (Baudais et al., 1997; Maier, 2001; Maier and Herbig, 2011; 84 85 Riehl, 2004). Sometimes, this type of sampling was combined with extensive coarse-sieving, which allowed observing some of the biases of small-volume samples (Maier, 2001). 86

87 The main reason why large-volume samples are rarely taken in wetland sites is that the

88 archaeobotanical evaluation of the samples is very time consuming. Furthermore large-

volume samples can pose problems in sites with very thick (superimposed) cultural layers

90 that might respond to more than one settlement phase, since these samples are difficult to

91 ascribe to a particular phase if this was not possible to identify during fieldwork (such a case 92 was observed at Pfäffikon-Burg in Zibulski, 2010). On a more practical scale, large samples also involve storage difficulties, since they need to be stored in cool dark rooms (or even 93 deep frozen) to avoid the degradation of the plant material present in them. Most sites where 94 95 large samples were investigated usually had to reduce the number of samples analysed (see 96 Fig. 1). Sites where small-volume samples were taken rarely reached 50 liters of sediment sieved in total. For this reason, the sampling strategy applied at Zürich-Parkhaus Opéra (with 97 98 ca. 1000 I of sediment processed) represents a milestone in archaeobotanical research in 99 prehistoric lakeshore research and can be used as a reference point to review previous 100 research.

- 101 The goals of this paper are:
- testing the comparability of the ubiquity, the concentration values (density values),
 the proportion (relative percentage) and the spatial analysis (using GIS) of large-sized
 items obtained in the 2mm fraction of subsamples of different volume taken from the
 same original sample;
- assessing which taxa are more often represented in the 2 mm and the 0.35 mm
 fraction in large and small-volume subsamples taken from the same samples;
- comparing the results of our test with those obtained from roughly contemporary
 investigated lakeshore settlements with different sampling strategies;
- 4. providing guidelines for the optimal procedures to efficiently record these plants in
 wetland sites and some final thoughts on the reliability of data obtained from samples
 of small volume (< 0.5 litres of sediment).



- 114 Fig. 1. Total volume of sediment (in litres) and number of samples sieved per settlement
- 115 phase of Neolithic lakeshore sites in the Alpine Foreland. Crosses refer to sites where the
- average volume per sample was above 0.9 L. Data compiled by S. Jacomet (ESM 1).
- 117

118 2. Materials and methods

119 Zürich-Parkhaus Opéra (Zürich, Switzerland) is a lakeshore site with several settlement 120 phases which was excavated during 2010 and 2011 (Bleicher and Harb, 2015). This paper 121 focuses on the methodological research carried out with samples from one settlement phase, 122 layer 13 (Horgen culture, dendrodated to c. 3160 BC, of ca. 20 years of duration (Bleicher 123 and Burger, 2015)). The sampling strategy applied at the site has been explained in previous 124 publications (Antolín et al., 2015; 2017; Steiner et al., 2015; Steiner et al., in press). Large-125 volume surface samples (5–7 I, the master samples) were taken in a systematic way. These 126 were subsampled before processing in the laboratory into two subsamples: the so-called A-127 and B-samples. B-samples were of smaller-volume (of 0.3 l), were taken using the grid 128 method taking sediment systematically from each square of the grid (see Steiner et al., in 129 press) and sieved with meshes of 2 and 0.35mm size. All B-samples yielded many more 130 remains than those that we aimed to recover for a representative evaluation of small-sized 131 items (ca. 400 remains, following Van der Veen and Fieller (1982) modified by Hosch and 132 Jacomet (2001)). For this reason, the 0.35mm fraction was always subsampled using the 133 grid method (like for master samples) and sub-samples of ca. 5 ml were analysed until the 134 target population was reached. In order to see what volume was necessary to sieve 135 concerning A-samples to recover a sufficient amount of large-sized remains (ca. 400) in the 136 2mm fraction, we performed an early evaluation of the data (unpublished). This volume was 137 observed to be around 3 litres. In consequence, A-samples were either the amount of 138 sediment that remained after the subsampling process to obtain the B-sample or, if this was 139 above 4 l of sediment, a subsample of it (always of above 3 l of volume). Therefore, A-140 samples usually had a larger volume of sediment (ca. 3-5 l). They were sieved with meshes 141 of 8 and 2 mm size. The wash-over technique with freezing as pre-treatment was used for 142 processing all samples (Kenward et al., 1980; Vandorpe and Jacomet, 2007). Over 250 A-143 samples and 120 B-samples were analysed completely. The 2 mm fraction was analysed in 144 both A- and B-samples in 96 samples (see location in Fig. 2). This allowed a unique 145 possibility to compare the results obtained in both. Nevertheless, since sediment was 146 removed from the original sample to obtain the B-sample, we cannot exclude a sequencing 147 effect in our test. We need to assume that since B-samples usually comprised less than 1/20 148 of the total amount of the master sample (of usually ca. 6 litres of sediment), the impact 149 should be of very low significance.

150 Quantification criteria were established in previous publications, so that certain remains were 151 only quantified in one of the fractions (Hosch and Jacomet, 2004; Steiner et al., 2015) (Fig. 152 3). The aim of this was to restrict the number of remains to be counted in the 2 mm fraction 153 to those taxa that are not found in the 0.35 mm fraction because of their larger size. This is 154 an efficient time-saving strategy in the analysis of the 2 mm fraction and it is based on the 155 assumption that the rare finding of smaller-size taxa in the 2 mm fraction (due to the sieving process, which does not perfectly separate both fractions) does not have a significant effect 156 157 on the overall results for the sample. Taxa that were not to be counted in A-samples (that is 158 to say, small-sized items) were described as present or absent in order to at least have their

- 159 presence recorded in the fraction. This was particularly of interest for those samples for
- 160 which no B-sample was analysed due to time and budgetary restrictions.
- 161
- 162



- 164 Fig. 2 Distribution of the points where both A- and B-samples from layer 13 at Zürich-
- 165 Parkhaus Opéra. In several cases, more than one sample from the same square was
- analysed, representing different parts of the inner stratigraphy of the cultural layer.



169 Fig. 3 Guidelines for the recording of botanical macroremains used in the analysis of

170 Parkhaus-Opéra (Steiner et al., 2015).

171 All sediment volume measurements presented in this paper refer to the displacement volume

172 (Antolín et al., 2015). Nomenclature of scientific plant names follows the National Data and

173 Information Center of the Swiss Flora (http://www.infoflora.ch).

174 We calculated the ubiquity values (number of samples in which one taxon was found) for

175 large- and small- sized items per fraction in both kinds of subsamples in order to compare

176 how samples of different volumes affect their representation in the record. The results of

177 large-sized items found in the 2 mm fraction of A- and B-samples were represented in

178 scatterplots with concentration values (number of remains per litre of sediment) and 179 proportions (relative frequency or percentage of a taxon in relation to the total of remains)

proportions (relative frequency or percentage of a taxon in relation to the total of remains ofthe sample) of objects counted in the 2 mm fraction in both kinds of subsamples. The

- 181 software R (R CORE TEAM, 2016) was used for this. In addition to this we integrated both
- 182 ubiquity and concentration of large-sized items by producing GIS Plans with ArcGis (ESRI,

183 2010) with the data obtained from both sets of subsamples. The size of the symbols was

- 184 established with the Jenks natural breaks classification method (Jenks and Caspall, 1971).
- 185 Anselin Local Morans I was used to calculate the clusters (Anselin, 1995).

186

187 3. Results

188 3.1. General results

Around 80.000 plant macroremains were found in the 8 and 2 mm fractions of the 96 A-

samples included in this evaluation (in total, 371.75 l of sediment, av. volume of sample 3,7l;

av. density: 209.8 r/l), and around 7.000 in the 2 mm fraction of the B-samples (29.38 l; av.

volume of sample around 0.3 l; av. density: 236.25 r/l). Over 45 taxa with large-sized

diaspores were recovered in both types of subsamples. Only 5 taxa were found in one of the sample types exclusively. *Prunus padus, Rhamnus cathartica* and *Crataegus laevigata* were

- 195 only recovered in A-samples, while *Tilia platyphyllos* and *Laserpitium siler* only in the B-
- 196 samples¹. Among the best-represented large-sized items in both types of samples one can
- 197 find Arctium sp., Corylus avellana, Galeopsis tetrahit, Linum usitatissimum (large capsule
- 198 fragments), *Malus sylvestris*, Maloideae (pericarp), *Prunus spinosa, Quercus* sp., *Rosa* sp.
- and *Viburnum lantana*. Common small- to medium-sized items found in both samples are
- 200 Najas intermedia/marina, Potentilla sp., Fragaria vesca, chaff remains of Triticum
- 201 aestivum/durum/turgidum (or T. "nudum"), Triticum dicoccon and Hordeum vulgare, as well
- as other taxa like *Chenopodium album, Polygonum persicaria* or *Sonchus asper.*
- 203

204 **3.2. Ubiquity**

All of the large-sized items were found in higher ubiquity values in A-samples (Table 1). For

- some taxa, the differences were not qualitatively significant, because they were present in
- 207 almost all samples. This would be the case of *Corylus avellana*, *Linum usitatissimum* (large
- 208 capsule fragments), *Malus sylvestris* and pericarp fragments of Maloideae. On the other
- 209 hand, large differences of above 30 samples (ca. 1/3 of the total) were found for other taxa
- 210 like Agrimonia eupatoria, Fallopia convolvulus, charred grains of Hordeum vulgare, Prunus
- 211 spinosa, Rosa sp. and Viburnum lantana.

212 Small-sized items were semi-quantified or only indicated as present in A-samples (see 213 section 2). We also compared the ubiquity values obtained in both types of subsamples to 214 check if this semiguantification provided relevant information (Table 2). It was observed in 215 several cases that taxa that were found in the 2 mm fraction of A-samples were more rarely 216 found in the 2 mm fraction of B-samples and, instead, they were mostly recovered in the 0.35 217 mm fraction. This is the case of many taxa like: Carex sp., rachis fragments of Hordeum 218 vulgare, charred seeds of Linum usitatissimum, seeds of Papaver somniferum, Valerianella 219 dentata, Brassica rapa, Chenopodium album, Malva sylvestris, Polygonum aviculare, 220 Polygonum persicaria and Ranunculus repens. Other (clearly small-sized) taxa were found 221 only rarely in the 2 mm fraction of both kinds of subsamples and mostly in the 0.35 mm 222 fraction of B-samples: Potentilla sp., Lycopus europaeus, seeds without wings of Betula 223 pendula/pubescens, among others. There were only a few cases of very abundant taxa 224 which were found to show similar ubiquity values in all fractions of all subsample types: 225 uncharred chaff remains of Hordeum vulgare, Triticum dicoccon, Triticum durum/turgidum 226 and seeds of Linum usitatissimum. Unexpectedly, a few other medium-sized items were more often found in A-samples. These include: Najas marina/intermedia (complete and half 227 228 seeds), charred chaff remains of Triticum dicoccon and Triticum durum/turgidum and seeds 229 of Aethusa cynapium as well as Ranunclus repens.

	A-Samples	B-Samples
Abies alba	32.3	11.5
Acer spec.	21.9	14.6
Agrimonia eupatoria	57.3	16.7
Alnus glutinosa, Catkin	11.5	2.1
Arctium spec.	94.8	68.8
Asteraceae, Flower	29.2	17.7

¹ We would like to note that these taxa were found in other A-samples of the same settlement phase not included in this evaluation.

Betula pendula/pubescens, Cone scale	43.8	25
Ceratophyllum demersum	14.6	1
Cerealia indet., Bran frag. (unch)	66.7	37.5
Cerealia indet., Grain (ch)	53.1	22.9
Clematis vitalba	36.5	9.4
Cornus sanguinea	26	5.2
Corylus avellana (ch)	31.3	8.3
Corylus avellana (unch)	100	88.5
Fagus sylvatica, Cupule	26	4.2
Fagus sylvatica, Pericarp	45.8	22.9
Fallopia convolvulus	57.3	20.8
Frangula alnus	16.7	2.1
Galeopsis tetrahit	81.3	52.1
Hordeum vulgare undiff., Grain (ch)	60.4	28.1
Linum usitatissimum, Capsule fr. (unch)	97.9	88.5
Malus sylvestris, seed	100	86.5
Malus/Pyrus, Pedicel (unch)	43.8	21.9
Malus/Pyrus, Pericarp (unch)	99	91.7
Malva sylvestris	26	2.1
Papaver somniferum, Capsule fr. (unch)	12.5	2.1
Prunus spinosa	86.5	53.1
Quercus spec.	52.1	24
Quercus spec., Pericarp (unch)	89.6	62.5
Rosa spec.	87.5	50
Sambucus nigra/racemosa	16.7	1
Triticum aestivum s.l./durum/turgidum, Grain (ch)	35.4	11.5
Triticum dicoccon, Grain (ch)	36.5	11.5
Viburnum lantana	79.2	32.3
Viscum album s.1.	25	14.6

Table 1. Ubiquity (percentage of samples) in which the large-sized items appear in A- and B-samples. Dark grey-shadowed taxa showed similarly high ubiquity values in both types of subsamples. If no indication is given, remains are preserved in an uncharred state.

			A-samples (2mm fraction)	B-samples (2mm fraction)	B-samples (0.35mm fraction)
Hordeum vulgare undiff.	Rachis segment	unch	72.9	59.4	66.7
Aethusa cynapium	Seed/fruit	unch	19.8	3.1	0
Alnus sp.	Seed/fruit	unch	19.8	0	16.7
Arenaria serpyllifolia agg.	Seed/fruit	unch	2.1	0	80.2
Betula pendula/pubescens, seeds with wings	Seed/fruit	unch	16.7	2.1	57.3
Betula pendula/pubescens, seeds without wings	Seed/fruit	unch	25	0	22.9
Brassica rapa	Seed/fruit	unch	17.7	0	22.9
Carex spec. bicarpellat	Seed/fruit	unch	11.5	3.1	19.8
Carex spec. tricarpellat	Seed/fruit	unch	40.6	4.2	52.1
Cerastium spec.	Seed/fruit	ch	1	0	1
Cerastium spec.	Seed/fruit	unch	1	0	40.6
Chenopodium album	Seed/fruit	unch	22.9	3.1	39.6
Fragaria vesca	Seed/fruit	unch	41.7	7.3	92.7
Hypericum perforatum	Seed/fruit	unch	2.1	2.1	1
Lapsana communis	Seed/fruit	unch	16.7	4.2	45.8
Linum usitatissimum	Seed/fruit	ch	18.8	2.1	15.6
Linum usitatissimum	Seed/fruit	unch	93.8	80.2	95.8
Lycopus europaeus s.l.	Seed/fruit	unch	14.6	2.1	44.8
Malva sylvestris	Seed/fruit	unch	26	2.1	13.5
Najas intermedia/marina	Seed/fruit	unch	77.1	58.3	35.4
Najas intermedia/marina	Fruit	unch	65.6	46.9	30.2
Origanum vulgare	Seed/fruit	unch	4.2	0	60.4
Papaver somniferum	Seed/fruit	unch	66.7	14.6	94.8
Physalis alkekengi	Seed/fruit	unch	63.5	14.6	66.7
Polygonum aviculare agg.	Seed/fruit	unch	37.5	0	59.4
Polygonum hydropiper	Seed/fruit	unch	16.7	3.1	13.5
Polygonum lapathifolium/persicaria	Seed/fruit	unch	2.1	3.1	11.5
Polygonum persicaria	Seed/fruit	unch	52.1	8.3	43.8

Seed/fruit	unch	14.6	4.2	81.3
Seed/fruit	unch	24	5.2	50
Seed/fruit	unch	62.5	11.5	43.8
Seed/fruit	unch	13.5	44.8	75
Seed/fruit	unch	87.5	15.6	68.8
Seed/fruit	unch	17.7	8.3	51
Seed/fruit	unch	10.4	3.1	24
Seed/fruit	unch	24	3.1	5.2
Seed/fruit	unch	33.3	7.3	25
Seed/fruit	unch	8.3	4.2	20.8
Seed/fruit	unch	25	3.1	65.6
Seed/fruit	unch	18.8	2.1	32.3
Seed/fruit	unch	9.4	2.1	5.2
Glume base	ch	30.2	6.3	7.3
Glume base	unch	85.4	74	84.4
Rachis segment	ch	17.7	9.4	6.3
Rachis segment	unch	76	67.7	85.4
Seed/fruit	unch	11.5	2.1	70.8
Seed/fruit	unch	15.6	4.2	21.9
Seed/fruit	unch	18.8	2.1	86.5
	Seed/fruit	Seed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchGlume basechGlume baseunchRachis segmentunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunchSeed/fruitunch	Seed/fruitunch14.6Seed/fruitunch24Seed/fruitunch62.5Seed/fruitunch13.5Seed/fruitunch87.5Seed/fruitunch17.7Seed/fruitunch10.4Seed/fruitunch33.3Seed/fruitunch87.5Seed/fruitunch10.4Seed/fruitunch10.4Seed/fruitunch33.3Seed/fruitunch8.3Seed/fruitunch25Seed/fruitunch18.8Seed/fruitunch9.4Glume basech30.2Glume baseunch85.4Rachis segmentch17.7Rachis segmentunch76Seed/fruitunch11.5Seed/fruitunch15.6Seed/fruitunch18.8	Seed/fruit unch 14.6 4.2 Seed/fruit unch 24 5.2 Seed/fruit unch 62.5 11.5 Seed/fruit unch 13.5 44.8 Seed/fruit unch 87.5 15.6 Seed/fruit unch 17.7 8.3 Seed/fruit unch 10.4 3.1 Seed/fruit unch 24 3.1 Seed/fruit unch 24 3.1 Seed/fruit unch 24 3.1 Seed/fruit unch 33.3 7.3 Seed/fruit unch 8.3 4.2 Seed/fruit unch 8.3 4.2 Seed/fruit unch 8.3 4.2 Seed/fruit unch 9.4 2.1 Glume base ch 30.2 6.3 Glume base unch 85.4 74 Rachis segment ch 17.7 9.4 Rachis segment

233 Table 2. Ubiquity (percentage of samples) in which the (small-sized) taxa that were semi-

234 quantified in A-samples appear in A- and B-samples (2mm and 0.35mm fractions

separately). Light grey-shadowed taxa showed higher ubiquity values in one of the fractions 235

or subsample types. *Rubus species were usually not identified to species level in the A-236 237 samples

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239 **3.3. Concentration values and proportions**

240 Differences between A- and B-samples in the mean concentration of large-sized items were not significant (Table 3). 241

	A-SAMPLES	B-SAMPLES
Hordeum vulgare undiff., Grain (ch)	1.8	2.2
Triticum aestivum s.l./durum/turgidum, Grain (ch)	0.4	0.6
Triticum dicoccon, Grain (ch)	0.4	0.4
Cerealia indet., Grain (ch)	1.0	0.9
Cerealia indet., Bran frag. (unch)	5.8	3.5
Linum usitatissimum, Capsule fr. (ch)	3.7	2.1
Linum usitatissimum, Capsule fr. (unch)	44.2	30.7
Papaver somniferum, Capsule fr. (unch)	0.1	0.1
Asteraceae (flower)	0.3	0.9
Abies alba	0.1	0.9
Acer spec.	0.1	0.5
Agrimonia eupatoria	0.3	0.6
Alnus glutinosa (catkin)	0.1	0.3
Arctium spec., seeds	5.9	8.1
Betula pendula/pubescens (cone scale)	0.4	1.2
Ceratophyllum demersum	0.1	0.0
Clematis vitalba	0.3	0.5
Cornus sanguinea	0.1	0.2
Corylus avellana (ch)	0.2	0.3

Corylus avellana (unch)	23.9	27.9
Fagus sylvatica, Cupule	0.1	0.1
Fagus sylvatica, Pericarp	1.8	1.7
Fallopia convolvulus	0.8	0.8
Frangula alnus	0.0	0.1
Malus sylvestris, seed	16.2	14.3
Malus/Pyrus, Pericarp (unch)	76.4	89.3
Malus/Pyrus, Pedicel (unch)	0.5	1.3
Malva sylvestris	0.4	0.1
Prunus spinosa	4.8	4.6
Quercus spec.	1.2	2.4
Quercus spec., Pericarp (unch)	13.1	18.8
Ranunculus repens	0.2	1.1
Rosa spec.	4.3	4.9
Sambucus nigra/racemosa	0.1	0.0
Viburnum lantana	1.0	1.4
Viscum album s.1.	0.1	0.8

Table 3. Mean concentrations of large-sized items in A- and B-samples (2mm fraction).

243 Scatterplots were produced in order to show the relationship (on a one-to-one basis)

244 between A- and B-samples concerning the density of the most-commonly-found taxa in A-

samples (Fig. 4). Secondly, the proportion of these taxa in relation to the total of the sample

246 were also plotted for comparison (ESM 2). All scatterplots are much skewed. In both cases

247 (Fig. 4 and ESM 2), at least two different patterns were observed. A few taxa yielded a better

distribution of a part of the samples along the line, indicating perfect match between both

subsample types, while a number of outliers is always present. This is the case of shell

250 fragments of *Corylus*, seeds of *Malus sylvestris*, pericarp fragments of Maloideae and large

capsule fragments of *Linum usitatissimum*. The rest of the taxa showed no clear pattern.



Fig. 4 Concentration values obtained for selected large-sized taxa in A- and B-samples. The red line shows perfect match

256

257

A Spearman's correlation was run to determine the relation between the values per taxon of A and B subsamples both using concentration values and proportions and a strong positive correlation was only determined for two taxa in both cases: *Agrimonia eupatoria* and *Ranunculus repens* (Table 4).

P: Spearman's Correlation	Concentration	Proportions
Triticum aestivum s.l./durum/turgidum, Grain (ch)	0.004	0.013
Triticum dicoccon, Grain (ch)	0.000	0.000
Cerealia indet., Grain (ch)	0.006	0.011
Cerealia indet., Bran frag. (unch)	0.052	0.162
Hordeum vulgare undiff., Grain (ch)	0.000	0.000
Linum usitatissimum, Capsule fr. (unch)	0.000	0.000
Agrimonia eupatoria	0.661	0.876
Arctium spec.	0.000	0.000

Corylus avellana (unch)	0.000	0.000
Galeopsis tetrahit	0.000	0.000
Malus sylvestris, seed	0.000	0.000
Malus/Pyrus, Pericarp (unch)	0.000	0.000
Prunus spinosa	0.000	0.000
Quercus spec., Pericarp (unch)	0.000	0.000
Ranunculus repens	0.748	0.641
Rosa spec.	0.000	0.000
Viburnum lantana	0.041	0.126

Table 4. Pearson's correlation, p-value. Strong correlations shaded in dark grey.

264

265 **3.3. GIS mapping of concentration values**

266 Taxa that were fully quantified in A- and B- samples were represented in GIS maps using the 267 Jenks natural breaks to define the size of the circles to avoid taking differences on a sample-268 to-sample basis, and focus on the trends observed of samples with "low", "medium" or "high" 269 density, and, in addition to this, pointing out statistically significant clusters to compare the 270 information provided by both datasets (see a summary of selected taxa in Fig. 5 and a more 271 complete list of taxa in ESM 2). For some large-seeded taxa, like pericarp fragments of 272 Maloideae, higher densities were found to be more widespread in the A-samples, and 273 therefore clustering was better identified (instead of outliers, which were detected in the B-274 samples). Clustering was also better identified for Arctium seeds and Viburnum lantana in 275 the A-samples. On the other hand, charred grain of *Triticum "nudum"* and large capsule

276 fragments of *Linum usitatissimum* yielded similar distributions and clustering patterns.

























Fig. 5. GIS plans for different taxa quantified in A- (left column) and B-samples (right column) showing the density of remains per sample using Jenks natural breaks to define symbol size and Anselin Local Morans I to identify clusters and outliers.

288

289 4. Discussion

290 **4.1. Limitations of this work**

The results presented in this paper are of high relevance for archaeobotany in wetland contexts, particularly taking into account the insufficient amount of methodological research done to date in this field, mostly due to the fact that most of the research is carried out in the framework of commercial archaeology, which does not allow the time nor the financial support for methodological evaluations.

296 The comparison presented above is aimed to make a final statement about the need of 297 large-volume samples from well-preserved waterlogged archaeological deposits in order to 298 have a full representation of all seed and fruit remains (possibly also other types of remains 299 recovered in these samples, like insect or fish remains). This was a necessary work given 300 that profile and core sampling are still very commonly used in similar contexts to the ones 301 investigated in this paper. This kind of sampling is most useful to target layer formation 302 processes, but not representative enough for a full reconstruction of the economy 303 (particularly regarding the relative importance of different plant foods in diet) at a site scale.

304 There are some methodological limitations in our work that need to be taken into 305 consideration. The samples we studied need to be understood as subsamples of different 306 volume of the same master sample, which is not fully comparable to the type of samples that 307 one would obtain by sampling profiles (monolith sampling), using cores scattered through the 308 site. At most, it could be compared to taking small surface samples because our samples 309 were taken from a surface of a square metre (so called scatter samples according to 310 Lennstrom & Hastorf 1992) and not just at a random point. One should assume that profile 311 sampling or core sampling would tend to give even more biased results than the ones 312 presented here, since the chances of not having a good representation of the whole surface 313 of the site are larger. In addition to this, taking a sub-sample from the master sample introduced a sequencing effect and an error (due to the process of subsampling a clumpy 314 315 sediment) that we have recently tried to calculate (Steiner et al., in press). Ideal case studies 316 or test studies do not generally exist in archaeology. One cannot take a large and a small 317 sample from a context without introducing this sequencing effect in some form. For this 318 reason we still find our analysis a powerful tool to judge the representativeness of different 319 sampling strategies in wetland sites with similar preservation conditions.

4.2. How comparable are ubiquity, concentration and proportions obtained in A- and B- samples for large-sized items?

323 We observed that the results obtained for large-sized items in A- and B-samples tend to 324 diverge considerably, depending on the method used to compare the samples. From the 325 most common taxa, only one was comparably well recorded in both kinds of subsamples: 326 large capsule fragments of *Linum usitatissimum* (Table 5). Some taxa never yielded 327 comparable results like charred grains, bran fragments of Cerealia or fruits of Agrimonia 328 eupatoria. Other plant macroremains, like pericarp fragments of Maloideae and nutshell 329 fragments of Corylus avellana gave roughly similar results for ubiquity, concentration and 330 proportions, but not in the GIS plans. Other taxa yielded similar GIS-plan distributions despite 331 dissimilarities in other parameters, like charred grains of Hordeum vulgare and Triticum 332 aestivum/durum/turgidum. Large-sized items seem to be systematically underrepresented in 333 small-sized samples except the most abundant and frequent ones (with the exception of 334 large capsule fragments of flax) (Steiner et al., 2015).

335 Regarding ubiquity values, on the basis of what we have observed (Table 1), very important 336 large-sized items present in almost all samples (97-100%) are equally recovered in almost all 337 smaller sub-samples (88-92%), but a few taxa with somewhat lower ubiquity values (75-338 95%), which are also economically important, presented clearly lower values in smaller 339 samples (30-60%), showing poorer chances to be detected with this type of sampling. 340 Therefore, under the pre-condition that a relatively large number of small-volume samples 341 have been investigated, large-sized items found in small-volume samples in ubiquities above 342 50% should be considered as important resources, since it is unlikely that these values are 343 higher for most taxa when small samples are taken. Ubiquity values are, therefore, in 344 general, not directly comparable to other sites where large-volume samples have been 345 investigated.

346 Regarding concentration, we have observed that average concentration values are similar 347 using both datasets and therefore are fully comparable, which is a very important result for 348 large-scale comparisons between different sites with different sample sizes. A relatively large 349 number of samples and a large-scale surface sampling (from multiple parts of the settlement) is also required in any case so that these average values are representative for the site. On 350 351 the other hand, comparisons on a sample-to-sample basis do not seem possible either 352 relying on concentration values or on proportions. Only two taxa (Ranunculus repens and 353 Agrimonia eupatoria) seemed to show a positive monotonic correlation, which is what one 354 would expect if B-samples provided a proportional amount of remains of those found in A-355 samples. In addition to this, a qualitative observation of the scatterplots in Fig. 4 and ESM 2 356 also allowed the observation that the taxa which have the highest ubiquity values in B-357 samples are the ones which also yielded more similar results in concentration on a one-to-358 one basis to A-samples. This could be interpreted as an indication that concentration values 359 at a sample scale are in general not reliable in small samples as a direct comparison to large 360 samples except for the most ubiguitous taxa.

Qualitative comparisons (combined with clustering analysis) between GIS plans produced
 with concentration values of A- and B-samples also showed divergences between A- and B samples, particularly among clearly large-sized items. For this reason, direct comparison
 does not seem possible.

	Ubiquity	Concentration	Proportion	GIS Plan
Malus sylvestris, seed				
Maloideae, pericarp fragment				
Corylus avellana, shell fragments				
Prunus spinosa, seed				
Rosa sp., seed				
Agrimonia eupatoria, fruit				
Cerealia, grain (charred)				
Cerealia, bran fragment (uncharred)				
Hordeum vulgare, grain (charred)				
Triticum "nudum", grain (charred)				
Linum usitatissimum, large capsule fragment				
Arctium sp., seed				

Table 5. Ticks mark an "acceptable" agreement in the results obtained in A- and B- samples.

367

368 **4.3. Which taxa appear in which fractions? Does sample volume make a difference?**

Two different aspects were to be considered regarding the type of remains found in each fraction: first of all, do large-sized remains appear in the 2 mm fractions independently of sample size? Secondly, are large- and small-sized items equally separated in the two fractions that we used in large and small samples? In consequence, were our counting units adequate for our case study and are they valid for other case studies with independence of the volume of the samples?

375 We had defined our **counting units** on the basis of previous work in Arbon Bleiche 3 (Hosch and Jacomet, 2004) and adapted them after performing some initial tests (Jacomet, 376 unpublished) and further sieving tests (Fig. 3, Steiner et al. 2015). With the present work, we 377 378 identified some other plant macroremains that were not quantified in the 2 mm fraction of A-379 samples but should be included in the quantification list in future analyses (these concern 380 mainly middle-sized seeds and other items): Najas marina/intermedia (complete and half 381 seeds), charred chaff remains of *Triticum dicoccon* and *Triticum durum/turgidum* and seeds 382 of Ranunculus repens and Aethusa cynapium (ESM 4). It is particularly important to emphasize that charred chaff remains of cereals must be counted in this fraction in order to 383 384 obtain comparable results to dry sites.

385

386 What about small-sized items? As already mentioned, smaller-sized items were semi-

387 quantified in A-samples. In the case that the 0.35 mm fraction had not been analysed, how

- much could one rely on the semiquantifications performed in A-samples? Could one use the
- 389 presence/absence of these taxa as a reliable indicator in, for instance, preliminary
- evaluations for a site under study? In this test, the recording of presence of the taxa proved
- to be reliable for a lot of taxa (including uncharred chaff remains of the main cereals and flax

- 392 seeds) except those with clearly small seeds like *Potentilla* sp., *Lycopus europaeus, Betula*
- 393 pendula/pubescens, Fragaria vesca, Origanum vulgare, Prunella vulgaris, etc. These taxa
- 394 would have always been underestimated in terms of ubiquity in the values produced by the 2
- 395 mm fraction of A-samples. One final remark is needed: our results are only valid for other
- 396 sites where the sieving method was equally gentle (the so-called wash-over technique).
- 397 Otherwise, much less small-sized material would have been recovered in A-samples.
- 398

399 4.4. Comparison with other contemporary lakeshore settlements

The results presented above indicate two main guidelines for the comparison of values regarding large-seeded taxa between sites where different sample sizes have been taken. The first one is that only the global concentration values (and not at a sample basis) can be compared if a relatively high number of samples has been studied. The second one is that only the most ubiquitous taxa will have a reliable ubiquity in sites where small samples were taken. Otherwise, taxa with ubiquities of 30-70% should be considered as very ubiquitous (=75-95%).

407 In order to see how useful these guidelines are, we compared our results with other roughly 408 contemporaneous lakeshore sites investigated in the region and for which the necessary 409 data were available: Arbon Bleiche 3 (Hosch and Jacomet, 2004), Horgen Scheller (Horgen layers) (Favre, 2002), Bad Buchau-Torwiesen II (Maier and Herbig, 2011) and Zürich-410 Kanalisationssanierung Seefeld (abbreviated as "KanSan") (Layer 3) (Brombacher and 411 Jacomet, 1997). These sites were sampled in different ways. From Arbon Bleiche 3 we could 412 413 include profile samples (av. volume of 0.13 litres per sample, 49 samples from 12 places 414 along a lake-land transect) and surface samples (33 samples of 2 litres of volume in 415 average); the latter came from different locations than the profile samples. From Horgen 416 Scheller, scatter samples (see section 4.1) of small volume (ca. 0.6 litres) were taken, while 417 from Zürich-KanSan only profile samples from 13 locations (ca. 0.7 litres of volume per 418 sample in average) were studied. We selected a number of taxa that appeared in our test study and, in order to avoid comparing very poor samples, only samples that presented more 419 420 than 100 remains per litre of sediment of these taxa were kept for the evaluation. The 421 synthesis of the data produced can be observed in Table 6.

422 It can soon be observed that the lowest density and ubiquity values were obtained mostly in 423 the profile samples of Arbon Bleiche 3 and the coring program at Torwiesen II, which also 424 presented the smallest volumes per sample, as well as in Horgen-Scheller, where the 425 average volume was below 1 litre per sample. It is particularly clear how charred cereal 426 remains were found in much lower ubiquities in all sets of samples except the surface 427 samples of Arbon Bleiche 3. These trends confirm the results observed in our test. The large 428 differences in concentration values between small- and large-volume samples observed in 429 Table 6 (particularly for Arbon Bleiche, where both types of samples were taken) might be 430 due to the fact that small-volume samples come from profile columns or cores (in Bad 431 Buchau). In both cases, the sample is taken from a specific point, instead of aiming to have 432 represented a larger surface as with scatter samples. As already noted in the comparison 433 performed by Jacomet between profile and surface samples at Arbon Bleiche 3, the average 434 concentration values of small-volume samples taken from a small number of spots (<20) 435 cannot provide reliable estimations (Jacomet et al. 2004, 413-414)

	-										1		
	Arbon B	leiche 3,	Bad Buchau-		Horgen-Scheller,		Horgen-Scheller,		Zürich-KanSan,				
	Profile	samples	Torwie	esen II,	Laver 3, s	Laver 3, surface		Laver 4, surface		profile column (18		Arbon Bleiche 3.	
	(49 sam	ples from	om coring program		samples (41		samples (21		Samples from 13		Surface samples		
	12 colu	mns, av.	(537 Samples, av.		samples, av. Vol. 0.6 samples, a		av. Vol. 0.7	columns, av. Vol. 0.7		(33 Samples, av.			
	Vol. 0.1	13 litres)	Vol. 0.2	Vol. 0.225 litres)		litres)		litres)		litres)		Vol. 2 litres)	
	averag		averag				averag				averag		
	е		е		average		е		average		е	ubiquit	
	density	ubiquity	density	ubiquity	density	ubiquity	density	ubiquity	density	ubiquity	density	y	
Malus sylvestris, seed	100.1	93.9	1.5	5	69.5	92.7	14.3	95.2	61.2	100	179.4	100	
Malus sylvestris, pericarp frag.	284.6	100	0.6	<1	363.3	100	37.8	100	124.5	100	220.1	100	

Quercus spec., pericarp frag.	5.0	26.5			79.2	95.1	32.0	100	104.0	77.8	16.1	87.9
Corylus avellana, shell frag.	183.0	95.9	0.1	<1	73.1	92.7	42.6	100	38.0	100	273.5	97.0
Prunus spinosa, stone	2.0	16.3	0.0	<1	2.7	53.7	3.8	47.6	6.5	83.3	2.9	75.8
Rosa spec., stone	2.3	14.3	0.0	<1	9.8	82.9	6.8	85.7	14.9	88.9	3.2	72.7
Cerealia indet., grain (ch)	3.2	14.3	0.0	<1	8.7	80.5	0.9	33.3			1.5	63.6
Hordeum vulgare undiff., grain (ch)	13.7	26.5	2.0	15.0	0.9	36.6	0.8	33.3	1.3	33.3	1.8	78.8
Triticum "nudum", grain (ch)	1.7	12.2	0.4	6.0	0.8	36.6	0.3	14.3	0.9	22.2	1.2	69.7
Linum usitatissimum, cansule frag	239.0	91.8	76.0	79	1014 1	100	2508.4	100	984.4	100	1136.7	100

Table 6. Average density and ubiquity of some selected large-seeded taxa from lakeshore
sites in the Alpine Foreland that are roughly contemporary to Zürich-Parkhaus Opéra,
ordered from left to right according to the mean sample volume. The lowest values per taxon
are shaded in grey. Volume measurements of Arbon Bleiche 3, Zürich-KanSan and HorgenScheller were converted to displacement volume by applying a factor of 1.5 as suggested in
Antolín et al., 2015. See references to the bibliographical sources in the text.

Regarding the number of places that should be sampled if only profile columns or cores with samples of less than 1 litre of volume in average are a viable option or in order to consider this type of approach as representative, 20 sampling spots have been mentioned in previous research (Jacomet et al. 2004, 414). We would now even suggest 40 spots as a minimum, taking into consideration the results of layer 3 in Horgen Scheller (Table 6). This number remains to be tested, since the work presented here does not allow stating a minimum number of samples.

449 450 450 451 452 453 454 454 455 455 455 456 457 457 457 458 459 459 450 450 450 451

The analysis of ubiquity showed that some plant macros were clearly underrepresented in samples of small size like charred cereal grains, and wild plants with a secondary importance (not regularly consumed and processed, but economically important) like *Viburnum lantana, Rosa* sp. or *Prunus spinosa*. In this case, only very frequent and abundant taxa like *Corylus avellana* or *Malus sylvestris* produced similar patterns for large- and small-volume samples.

457 On the other hand, the mean concentration per taxon seems to not be significantly different 458 between subsample types, which is probably not comparable to sites where profile or core 459 samples were taken because these only reflect very specific areas of the surface of the site. 460 Therefore, if a large number of small samples (above 40) is taken and the spatial patterning 461 of each plant taxon is not a main question of the project, a reliable average density can be 462 obtained for a settlement phase and reliable ubiquity values for all those taxa that appear in 463 high densities and extremely high ubiquities. For large-sized taxa that do not appear in most 464 samples and particularly taxa that appear in low densities, small-sample volumes are not appropriate. This implies, for instance, that charred grains (probably excluding those in burnt 465 466 layers) were not representatively recorded in sites where only samples of small volume were 467 taken.

From the results obtained at Zürich-Parkhaus Opéra, we can infer that samples of 3 l of sediment volume (that is to say 4.5 l if measuring the classical volume) are needed for a representative recovery of large-sized plant remains. This confirms the estimations done in previous work, but tests should be carried out in other sites to prove the applicability of this value (it can vary if this amount is not enough to recover ca. 400 large-sized items, for instance).

- 474 This result implies that more human and financial efforts need to be put in the
- 475 archaeobotanical research of sites with waterlogged preservation, and that more

- 476 methodological critique needs to be included in the interpretation of archaeobotanical
- 477 assemblages coming from samples of small volume in this type of archaeological deposits.
- 478

479 **5. Conclusions**

480 In this study we could perform a test analysis that finally proves that large-volume samples are needed for the representative recording of large-sized items (including charred cereal 481 482 grain) in wetland sites, especially if the economy of a site is targeted. This is an important 483 result given that sample sizes are usually very small (below 1 litre) in these sites, which 484 renders the results of these unique contexts (regarding charred remains) almost 485 incomparable to the archaeobotanical data obtained from dry sites. We therefore recommend 486 the use of samples of around 3 I of sediment, at least, for well-preserved Neolithic settlement 487 layers (with average concentration values per sample of around 10.000 mostly uncharred 488 remains per litre of sediment).

- 489 We could also show that average density values obtained from small-volume samples might
- 490 be comparable to those obtained from large-volume samples. This facilitates inter-site
- 491 comparisons of wetland sites, although caution is always recommended in interpretations.
- 492 Ubiquity values of large-sized items will probably be underestimated in sampling programs
- based on samples of small volume. In this case, ubiquity values above 30% and particularly
 above 50%, need to be considered as very high. Site plan distributions of large-sized items
- above 50%, need to be considered as very high. Site plan distributions of large-sized items
 based on samples of small volume are prone to a large bias and should not be performed or
- 496 interpreted as representative (particularly the absence of remains cannot be interpreted in
 497 these cases).
- Since the number of remains to be counted per sample are independently treated for each
 fraction (ca.400 remains are needed per fraction), counting units need to be clearly predefined to avoid counting remains twice and our proposal can be used as a reference. A
- 501 proposal is included in the ESM 4 of this paper.
- 502 Finally, we recommend the conduction of tests at a site scale to check the suitability of these 503 standards to each case study.
- 504

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