

# THE DOG OR ITS MASTER? AN INTERDISCIPLINARY STUDY ON COPROLITES FROM THE PILE-DWELLING SITE STARE GMAJNE\*

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## ABSTRACT

The initial results on the seven Late Neolithic dog coprolites from Slovenia discovered in 2007 and 2014, and published in 2019 and 2021 are now complemented by the new results on two newly discovered coprolites in 2021. The pollen, plant and animal macro-remains, as well as parasites preserved indicate that we are probably dealing with two different origins, both, the dog's and its master. The differences are presented.

## KEYWORDS

coprolites, dog, human, Eneolithic, pile-dwellings, Ljubljansko barje, Slovenia / koproliti, pes, človek, eneolitik, kolišča, Ljubljansko barje, Slovenija

## INTRODUCTION

Coprolites are unique sources of diverse information for biology, ecology and archaeology.<sup>1</sup> The faecal biomarkers can help us to confirm the producer, digestive physiology, as well as the diet: plant (e.g. sitosterols present) vs. animal (e.g. cholesterol present). The seasonality of the defecation could be confirmed additionally with plant macro-remains: i.e. summer fall vs. winter/spring fall plant taxa.<sup>2</sup> The origin of the coprolite is first determined by morphology, i.e. colour, outer structure, shape and the size. Dog excrements appear very similar to that of humans, but the inner composition may differ. Numerous digested bone fragments are commonly preserved in the dog coprolites, mainly fish bones, in contrast to human faeces which contain mainly plant remains, especially of cereals.<sup>3</sup> On the other hand recent research shows doubts on such a simple separation of the origins, not only because human and dog excrements are often similar in size and shape, they tend to occur together at archaeological site. Moreover, dogs often consume food similar to those of humans and are even consuming human food and their faecal remains.<sup>4</sup> Anyway, the size of the bone fragments (less than 5 mm) and the presence of medicinal plant macro-remains, like e.g. white mistletoe and ivy, could confirm that it is more likely human excrement. Especially plant taxa that treat respiratory diseases and with antiparasitic effect, i.e. against diarrhoea and abdominal pain, as well as taxa with diuretic properties, may be crucial in determining human origins.<sup>5</sup>

The excavations at two Eneolithic pile-dwelling sites (Črnelnik and Stare gmajne) at Ljubljansko barje in Slovenia, yielded sub-fossil excrements (i.e. coprolites; Image 1). They were of likely-human or -dog shape and size. Their content, which was analysed, showed that we were dealing with the coprolites of a dog origin.<sup>6</sup> Palynological, palaeoparasitological, archaeobotanical and archaeozoological features investigated, provided also some additional informations, such as health, status, nutritional habits, environment conditions and the season, beside the diet.

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\* Translation: dr. Tjaša Tolar

<sup>1</sup> e.g. REINHARD, K. J., BRYANT, V. M. 1992; WOOD, JR., WILMSHURST, J. M. 2016.

<sup>2</sup> e.g. JAKOBITSCH, T. et al. 2023.

<sup>3</sup> BYRNE, D. 1973; BRÖNNIMANN, D. et al. 2017; TOLAR, T., GALIK, A. 2019; TOLAR, T. et al. 2021.

<sup>4</sup> KUBIAK-MARTENS, L. 2022a, p. 31.

<sup>5</sup> e.g. KUBIAK-MARTENS, L. et al. 2022, pp. 135-137.

<sup>6</sup> TOLAR, T., GALIK, A. 2019; TOLAR, T. et al. 2021.

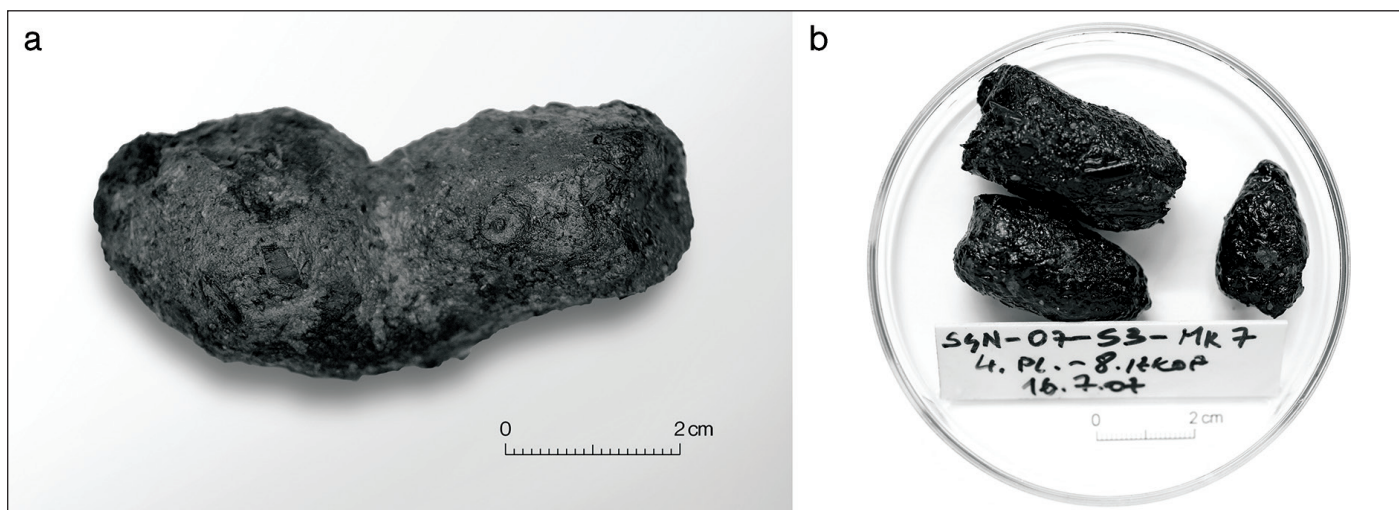


Image 1: The coprolites from a) Črnelnik site and b) Stare gmajne 2007 site. Photo: D. Valoh, IZA ZRC SAZU.

The first analysis of coprolite in Slovenia was done on the one, found in 2014 at Črnelnik, an Eneolithic pile-dwelling site (Image 1a). It was very well preserved and only macro-analysis was performed as this was the first such investigation in Slovenia. Its content was mainly of fish skull bones, in addition to some pharyngeal teeth and scales of Cyprinidae fish family. Rare plant macro-remains were found in it as well, i.e. 4 blackberry seeds, 3 white goosefoot seeds, 1 flax, turnip and birch seed and 2 fragments of the water chestnut fruit and leaf. They confirm the autumn time of defecation.<sup>7</sup> Besides the evidence of feeding only with fish heads, which is not common for humans, the absence of larger proportion of plant remains, especially of cereals and gathered fruits (like raspberries, strawberries, apples, etc.), testify that the coprolite from Črnelnik site originates from a dog, rather than human. A variety of nuts, fruits, and berries were collected and processed for food, as indicated by their macro-remains in the cultural layers of the Eneolithic pile-dwellings.<sup>8</sup> They were probably deposited as a remnant of food supplies and food preparation in the settlement, and may also be a remnant of human and domestic animal excreta. Blackberry, raspberry, strawberry, elderberry seeds, for example, pass through the digestive tract and are excreted intact. Whole human/dog coprolites in its original form are rarely found at archaeological sites, although faeces were deposited together with other sediment during the existence of the settlement, but could often not be distinguished. Therefore a mixture of all is most likely the material of the cultural layer we are usually dealing with.

The second survey of coprolites in Slovenia was conducted on six coprolites from another 4<sup>th</sup> mill. BC pile-dwelling site, i.e. Stare gmajne site (Image 1b). Altogether 16 coprolites were found in a 15 m<sup>2</sup> trench 3 (Image 2) during excavation in 2007. They were kept in distilled water and under cold condition (i.e. 4°C). Five different analyses were performed on six coprolites (palynological, palaeoparasitological, archaeobotanical and archaeozoological). However, not all analyses were possible on all six examined coprolites, since there was not enough material (i.e. 3 – 20 ml) available. Among four of the macro-analysed coprolites, all contained fish vertebrae and flat cranial fish bones. Three of them contained also digested mammal and bird bones. Two of them contained fish pharyngeal teeth. Other fish remains, like fish scales, fin rays and ribs were obtained only in one of the investigated coprolite.<sup>9</sup> Plant macro-remains (seeds, fruits frg.) were found in three out of four investigated coprolites, i.e. of cereal, flax, white goosefoot and lakeshore bulrush, but with very low abundance (i.e. 1-2, rarely 5 items/fragments per a coprolite) and not all ID taxa were present in all investigated samples.<sup>10</sup> The results of macro-remains in all four coprolites from Stare gmajne site indicate, similar to the Črnelnik coprolite, minor consumption of plants and major consumption of small vertebrate animals, mostly fishes. Apparently we were dealing with the dog excrements again, which was also confirmed by the palaeoparasitological analyses. In all of the three parasitologically analysed coprolites, eggs of intestinal par-

<sup>7</sup> TOLAR, T., GALIK, A. 2019.

<sup>8</sup> TOLAR, T. et al. 2011; TOLAR, T. 2018.

<sup>9</sup> TOLAR, T. et al. 2021, p. 111.

<sup>10</sup> TOLAR, T. et al. 2021, p. 110.

asites were identified. They were of three species of flatworms and whipworms, which all can be transmitted to mammals through consumption of raw or undercooked fish or meat of various animals. Particularly the presence of *Alaria*, fluke genus (common for wild and domestic canids), which was found in one coprolite in greater numbers, confirms a canid origin.<sup>11</sup> One coprolite from Stare gmajne was palynologically tested, the result shows high number of water-dependent plant taxa, especially Cyperaceae. Anthropogenic indicators, like cereals were present in minor percentages, i.e. less than 1 %. Outstanding presence of pollen of early successional taxa (i.e. birch, hazel, alder) indicate an early spring deposit period.<sup>12</sup>

To summarize, all, Črnelnik and 6 of the Stare gmajne 2007 coprolites, probably originate from the dog. They were all found in the cultural layer. The dogs were mainly fed by the remains of fish, probably caught by men and were drinking water from the nearby lake or river. The coprolites also give information about the defecation period, i.e. spring in the case of one coprolite from Stare gmajne<sup>13</sup> and autumn in the case of coprolite from Črnelnik site.<sup>14</sup> The dogs were infected by intestinal parasites, tapeworms, flukes and whipworms.

Due to the good research potential and high abundance of the coprolites at Stare Gmajne site, we decided to excavate again in the vicinity of the trench 3 (in 2007). Beside great potential for recovering new coprolites at the site, also additional research can be conducted on the freshly excavated coprolites. New excavation and investigation was held in the summer of 2021 as a part of the project “*Dog or its master? The scientific study of human or canine coprolites from the prehistoric pile-dwelling site of Stare gmajne, Slovenia*” (J7-2598, project leader dr. Tjaša Tolar). The project has been funded by the Slovenian Research and Innovation Agency and the Fund for the Promotion of Scientific Research (Republic of Austria). Partners from foreign institutions (Austrian Academy of Sciences, Vienna University of Technology and University of Bourgogne Franche-Comte (France)) have been involved. In case new coprolites would have been found, new types of analyses would have been performed (e.g. biochemical analyses). 9 coprolites have been found in new excavated 8 m<sup>2</sup> trench 4 in 2021 at Stare gmajne. They are currently being analysed. Preliminary results on two of them are presented in this study. All of them were found in the cultural layer which consists of dark brown clayey silt and is located about 60 cm below the surface. Most of the archeological material (pottery, bioarchaeological, wooden and stone artefacts) was found in this layer. Northern part of the trench 4 was the thickest, i.e. 64 cm, and the thinnest was 24 cm in the southern part.<sup>15</sup>

All together 25 coprolites (i.e. 16 in trench 3 (2007) and 9 in trench 4 (2021)) have been found so far at Stare gmajne site (ca. 3300 cal BC). We have obviously come across the location of the defecation in the vicinity of the two excavated trenches (together of 23 m<sup>2</sup>), as it is typical for dogs to defecate often in the same place. The newest archaeobotanical investigation also suggests that the 2021 trenches 4, 5 (Image 2) are located very close to or even at the very edge of the 4<sup>th</sup> mill. BC pile-dwelling settlement.<sup>16</sup>

## MATERIAL AND METHODS

Southeast to trench 3 (2007), two trenches were opened at Stare gmajne in 2021 (i.e. trench 4 and 5; Image 2). The excavation yielded 9 more likely-dog/human shaped coprolites, all from the trench 4. They were well preserved, some of them highly mineralized (i.e. Image 3c). The majority (7) were found in the two (A4 and B4) quadrants of the trench 4, which were closest to the trench 3 (see Image 2).

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<sup>11</sup> TOLAR, T. et al. 2021, p. 112, 114.

<sup>12</sup> TOLAR, T. et al. 2021, p. 114, 116.

<sup>13</sup> TOLAR, T. et al. 2021.

<sup>14</sup> TOLAR, T., GALIK, A. 2019.

<sup>15</sup> LEGHISSA, E. et al. 2022.

<sup>16</sup> TOLAR, T., MATIKA, D. in press.

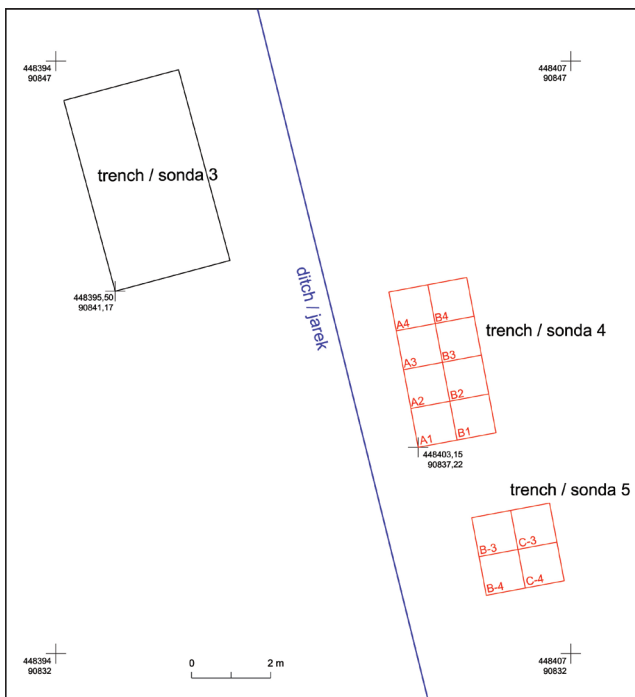


Image 2: The positions of the trenches 3 (year 2007) and 4, 5 (year 2021) at Stare gmajne site. Drawing: E. Leghissa and D. Valoh, IZA ZRC SAZU.

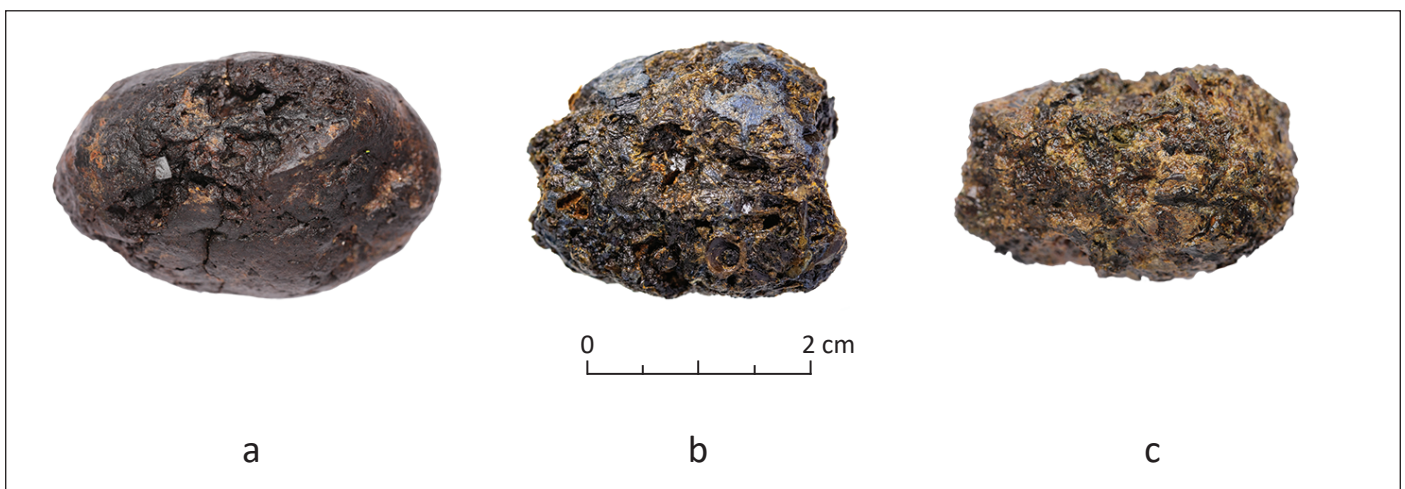


Image 3: Three examples of the coprolites found in trench 4 during the 2021 excavations at Stare gmajne site. In the middle (b) is the coprolite C2, discussed in this paper. Photo: D. Valoh, IZA ZRC SAZU.

Three samples were preliminary analysed and are presented here: two coprolites (C1 and C2) and one sediment sample (S3), that was deliberately taken just below the coprolite C2 (Image 3b), which we have found in the cultural layer during the excavation. Coprolite C1 was found during wet sieving and washing the vessel fragment taken from the field.

For all three samples archaeozoological, archeobotanical and palaeoparasitological analyses have already been done, while biochemical analyses are still in progress. One of them (i.e. C2) was also palynologically analysed.

For macro-analyses each sample was fine wet-sieved through 0.056 mm sieve mesh-size. The remains, caught on the sieve were waterlogged examined and sorted under stereomicroscope Leica with up to 50x magnification. For the identification the reference collection of plant and animal remains at the Institute of Archaeology ZRC SAZU as well as special literature<sup>17</sup> were used.

1 cm<sup>3</sup> of the subsample from the interior of the coprolite C2 was extracted for standard pollen analysis procedure (7% HCl, 10% NaOH, 40% HF, acetolysis, staining with safranine, mounting in silicone oil).<sup>18</sup> A Nikon

<sup>17</sup> e.g. CAPPERS, R. et al. 2006; JACOMET, S. 2006.

<sup>18</sup> BENNETT, K. D., WILLIS, K. J. 2002.

Eclipse Ci light microscope at 400x magnification, reference collection at the Insitute of Archaeology ZRC SAZU and identification keys were used for pollen identification.<sup>19</sup>

For parasitological analysis three samples were analyzed, corresponding to two coprolite samples (C1, C2) and a sediment sample (S3) taken under a coprolite. The samples were prepared following the standard RHM protocol (Rehydration-Homogenization-Microsieving) used in Besançon (France) to extract gastrointestinal markers.<sup>20</sup> Five grams of each sample were rehydrated for one week in 0.5% tri-sodium phosphate (TSP) and 5% glycinerated water. The samples were then crushed in a mortar and subjected to an ultrasound bath for one minute. Finally, the sediments were filtered in a column composed of sieves with 315, 160, 50 and 25 µm meshes. As gastrointestinal egg size varies between 30-160 µm in length and 15-90 µm in width, residues from the last two sieves were analysed.<sup>21</sup> Six slides were prepared from each fraction and observed under a light microscope. Identification of the taxa is based on morphological and morphometrical characteristics of the parasites eggs.

## RESULTS

### Plant and animal macro-remains analyses

In Tab. 1 the results of plant and animal macro-remains are given. The material of the two coprolites (C1, C2) is scarce, i.e. of only up to 5 ml of the wet material caught on the finest sieve. The sediment sample (S3) taken below the coprolite C2 (i.e. C2 was the only one found during the excavations) was larger (70 ml sediment sample). After wet-sieving, whole sample of 10 ml of the finest fraction S3 was examined.

Table 1: Macro-remains analyses in two coprolites C1 and C2 and in one sediment sample S3, taken below C2.

		C 1	C 2	S 3
plant taxa /remain / volume of wet fraction analysed		less than 5ml	5 ml	10 ml
<i>Triticum mono/dicoccum</i>	glume base C	2		2
<i>Triticum dicoccum</i>	spikelet fork NC	1		
<i>T. monococum</i>	spikelet fork NC			1
	spikelet fork C			1
<i>Linum usitatissimum</i>	capsule frg. NC			1
<i>Papaver somniferum</i>	seed NC			18
cf. <i>Brassica rapa</i>	seed NC			2
Maloideae	seed frg. NC	10		1
	pericarp frg. NC	1		1
	base NC	1		
<i>Fragaria vesca</i>	seed NC			6
<i>Rubus</i> sp.	seed frg. NC			1
cf. <i>Physalis</i>	seed frg. NC			1
cf. <i>Viscum</i>	fruit frg. NC	1		
<i>Corylus avellana</i>	fruit base frg. NC			1
<i>Trapa natans</i>	fruit base frg. NC			1
	leaf frg. NC			1
<i>Quercus</i> sp.	pericarp frg. NC	1	2	2
<i>Urtica dioica</i>	seed NC			4
<i>Chenopodium album</i>	seed NC	1 frg.	1 frg.	23
<i>Plantago</i> sp.	seed NC			1
<i>Oenanthe aquatica</i>	seed NC	1		9
<i>Schoenoplectus lacustris</i>	seed NC	1		6
<i>Cladium</i> sp.	seed NC			1
<i>Ranunculus aquatilis</i>	seed NC			8
<i>Mentha aquatica</i>	seed NC			6
<i>Carex</i> sp.	seed NC			1

<sup>19</sup> MOORE, P. D. et al. 1991; REILLE, M. 1992, 1995.

<sup>20</sup> DUFOUR, B., LE BAILLY, M. 2013.

<sup>21</sup> ASH, L. R., ORIHIEL, T. C. 2007.

Table 1: Macro-remains analyses in two coprolites C1 and C2 and in one sediment sample S3, taken below C2 (*continued*).

		C 1	C 2	S 3
<i>Potamogeton sp.</i>	seed NC			1
<i>Alnus glutinosa</i>	seed NC			3
<i>Abies alba</i>	needles frg. NC	7		3
mosses	NC			x
charcoal	frg. C	x	x	xx
wood, bark	frg. NC		4	x
amorphous mineralized frg. of food eaten	poss. scales, bones or plant remains mineralized into aggregates	xxx	x	
fish	scales frg. NC	xxx	x	xxx
fish	vertebra		6	x
fish	teeth		5	6
non-fish bones	digested bones frg. NC		xxx	
small mamal coprolites	NC			4
mollusca	frg. shells NC	1		5
insecta	frg. NC			x

frg. – fragment; x – present; xx – more; xxx – a lot  
C – carbonised; NC – not carbonised

All of them (C1, C2 and S3) contain fish remains, while the remains of other animals (i.e. bones of birds, mammals) could be determined only in C2. The fish vertebrae and teeth are present in C2 and S3, but not in the C1. Additionally, very little plant macro-remains were found in C2 (i.e. 3 fragments). In contrast, the absence of recognizable bones and teeth, and higher proportion and diversity of plant macro-remains (thus of cereals, apples and fir – i.e. cultivated and gathered plant taxa) were recognized in C1. It has to be mentioned here, that C1 coprolite had extremely mineralized matrix, with rough surface (see in the Tab. 1 – *amorphous mineralized frg.*), and it was therefore hard (or impossible) to wet sieve. The aggregates of mineralized food remains were strongly encrusted (see, e.g., Image 3c), which is not the case with all other examined coprolites from Slovenia (i.e., they were water-saturated, with fine, soft, and fragrant matrix that can be easily wet-sieved). Lots of fish scales in C1 absolutely indicates fish food.

The sediment below the C2 contain common plant macro-remains that are often found in the cultural layer of the pile-dwellings.<sup>22</sup>

### Pollen analysis

Pollen analysis was initially done on coprolite C2 (see Image 3b). 322 pollen grains were found and identified. The concentration of pollen was low (i.e. 8,893 pollen grains/cm<sup>3</sup>).

Table 2: Pollen analysis in coprolite C2, with absolute number of counted pollen grains.

<i>Pinus</i>	4	<i>Alnus</i>	22	Asteraceae	1
<i>Picea</i>	15	<i>Corylus</i>	22	Umbelliferae	1
<i>Abies</i>	2	Rhamnus	1	Filicales	4
<i>Larix</i>	1	Cyperaceae	184	Trilete spore	1
<i>Betula</i>	1	Poaceae	9	<i>Thelypteris palustris</i>	10
<i>Fagus</i>	4	Cereals	13	<i>Selaginella</i>	1
<i>Tilia</i>	5	<i>Rumex</i>	1	<i>Iris</i>	1
<i>Quercus</i>	15	<i>Ranunculus sp.</i>	2	<i>Typha</i>	1
<i>Carpinus betulus</i>	1				

<sup>22</sup> TOLAR, T. et al. 2011; TOLAR, T. 2018.

Palynological analysis suggests that vegetation around Stare gmajne mostly consisted of water-dependent taxa (*Alnus*, *Thelypteris palustris*, *Typha* and especially high percentage of Cyperaceae). Large percentage of *Quercus*, *Corylus*, *Alnus* and low percentage of *Fagus* and *Abies* would suggest that the landscape was quite open. Percentage of cereal grains (type Cerealia) represent 4% of the total pollen sum. The proportion of herbal taxa is small, only individual grains of *Rumex* sp., Umbelliferae, Asteraceae were found, which indicates local absence of grasslands. The pollen assemblage in C2 indicates spring to early summer deposit period.

### Palaeoparasitological analysis

All tree samples (C1, C2, S3) were palaeoparasitologically tested. Three taxa of gastrointestinal parasites of humans or animals were identified during our analyses (Tab. 3). They belong to three classes of intestinal worms: the Nematodes, the Trematodes and the Cestodes. Because the samples were taken from an occupation layer, their biological origin cannot be specified for the moment. In these conditions, the determination of the parasite eggs remains difficult and often cannot reach the species level.

Table 3: Identified eggs of the intestinal worms in three samples of 5 grams from Stare gmajne 2021 (trench 4): C1, C2, S3.

			C1	C2	S3
whipworms	<i>Trichuris</i>	Nematodes	39	1	18
tapeworms	<i>Diphyllobothrium</i>	Cestodes	1		3
flukes	<i>Echinostoma</i>	Trematodes	1		2

Eggs belonging to the genus *Trichuris* (whipworm) were identified and found in high proportions in one sample (i.e. C1). The eggs of *Trichuris* are the most frequently found in the archaeological samples,<sup>23</sup> the result of S3 (i.e. sediment sample of the cultural layer under the C2) could prove this statement. It is a cosmopolite and ubiquitous parasite affecting a large number of species including humans. In the present case, the egg average size of 55 x 30 µm allowed us to attribute them to the human species (*T. trichiura*) or the pork species (*T. suis*). The coprolite C1 contain eggs of *Trichuris* genus in greater number than C2 and has a similar parasite spectra as S3 – the sediment sample from the cultural layer, i.e. under C2.

## DISCUSSION

Coprolite is a term which encompasses faecal material preserved either by mineralization or desiccation, from both paleontological and archaeological contexts.<sup>24</sup> In general, coprolites are usually preserved by mineralization, but exceptions are pile-dwelling sites in Europe where coprolites are usually preserved in waterlogged conditions.<sup>25</sup> In the case of Stare gmajne (excavations 2021) nine likely human/dog or even pig coprolites were found in trench 4. Both coprolites that were analysed showed different morphological appearances. C1 had extremely mineralized matrix and rough surface which made wet sieving and recognizing macro-remains difficult. On the other hand coprolite C2 was very similar to the firstly analysed coprolites (i.e. from Črnelnik and Stare gmajne 2007), water-saturated and with fine, soft, and fragrant matrix that was easily wet-sieved and macro-remains sorted out. Whether the circumstances of the environment where the investigated coprolites were preserved differ or this is the consequence of different origin of the excrement (i.e. dog, human, pig) is hard to discuss. Biomineralization and the precipitation of calcium carbonate is quite normal and often at archaeological sites, especially on manure pits, garbage dumps, septic tanks or cloacas, where increased amounts of organic compounds turn into dead inorganic compounds in the process of mineralization, for which it has a consequence that different remains are preserved differently under such circumstances.<sup>26</sup> The non-fine matrix, described for C1, has much similarities to the coprolites from the e.g. Late Neolithic settlements in the Netherlands, where the bones were embedded in the coprolite matrix as well. Among 16 multiproxy analysed coprolites from the Netherlands, 2 were determined for pigs, 3 for dog/human, 3 for human and 3 for pig/human mainly on the basis of faecal steroids (i.e. gas chromatography-mass spectrometry (GC-MS)).<sup>27</sup>

<sup>23</sup> BOUCHET, F. et al. 2003; GONÇALVES, M. L. C. et al. 2003.

<sup>24</sup> REINHARD, K. J., BRYANT, V. M. 1992.

<sup>25</sup> e.g. KUBIAK-MARTENS, L. 2022a, p. 31.

<sup>26</sup> WILKINSON, K., STEVENS, C. 2003; JACOMET, S. 2007.

<sup>27</sup> KUBIAK-MARTENS, L. et al. 2022, pp. 122-127.

Although only initially results at Stare gmajne 2021 (trench 4), the differences between the investigated samples can be obtained. Especially the coprolite samples C1 and C2 show a great difference in content of plant vs. animal macro-remains as well as in containing obviously more/less *Trichuris* (whipworm) eggs.

Table 4: Comparison of the analyses performed on C1, C2 and S3.

	C1	C2	S3
ARCHAEOZOOLOGY	fish scales	digested bones, fish vertebrae, teeth, scales	fish vertebrae, teeth
ARCHAEOBOTANY	27 ID items; thus: <i>Triticum</i> , <i>Malus</i> , <i>Abies</i>	minor extent, only 3 ID frg. of seeds/fruits	greater extent, 106 ID items
PALYNOLOGY	n.d.	Cerealia, Cyperaceae, <i>Thelypteris</i> , <i>Picea</i>	n.d.
PALAEOPARASITOLOGY	more <i>Trichuris</i>	only 1 <i>Trichuris</i>	more <i>Trichuris</i>

Sample S3 contains quite the same remains as they are often found in the cultural layers of the pile-dwellings,<sup>28</sup> therefore we can attribute it as a sediment sample from the cultural layer. The high abundance of *Trichuris* eggs can probably confirm this assumption, while the eggs of *Trichuris* are most frequently found in the archaeological samples.<sup>29</sup> It is a cosmopolite and ubiquitous parasite affecting a large number of species including humans. The eggs of all species of *Trichuris* genus strongly resemble each other and sometimes show also very similar size distributions. In the presented case, the egg average size of 55 x 30 µm allowed us to attribute them to the human whipworm species (*T. trichiura*) or the pork species (*T. suis*). Infection occurs by ingesting mature eggs in polluted soils, water or food, and is related to bad body hygiene and inappropriate waste management. The use of faecal matter for crop fertilization contributes to maintain and amplify the parasite presence. The infection is usually asymptomatic. However, symptoms of tiredness, anemia or colitic disorders are possible in case of a strong infection. In future, it would be worth to palaeoparasitologically test the cultural layer at several places of the excavated area in order to find out how was the soil/sediment in the settlement itself polluted with those parasites.

### Coprolite C1

Coprolite C1, which was found in the ceramic vessel fragment during wet sieving and washing, contained more human- than dog-like indicators: no bones, fish scales (which could be eaten together with fish skin), and quite a lot of plant macro-remains (carbonised and non-carbonised cereal chaff fragments, different apple remains, acorn, fir needle, etc.). The greater presence of amorphous mineralized fragments compared to the other two analyzed samples (C2, S3; see Tab. 1) could be of taphonomical reason (i.e. mineralization; see the explanation above).<sup>30</sup> High proportion of *Trichuris* sp. (Nematodes; whipworm) eggs (39) confirm the human or pig origin of the coprolite C1 (see the description and detailed explanation above). As both, humans and pigs are omnivores, the macro-remains in the coprolite C1 could neither prove the exact origin. Mistletoe fragment, found in C1, could possibly be the remain of animal (pig) fodder,<sup>31</sup> but again it could be found in human coprolites as well, interpreted as medicinal plant.<sup>32</sup> The evidence for the cereal presence also can not be the prove for the origin (human/pig), while cereals were consumed quite often and the leftovers were probably fed to the animals. The fact that the coprolite C1 was found inside the settlement (possibly even in a house) and in a vessel, perhaps makes interpretation easier? It is probably of human origin, since pigs were presumably not present in the house, dogs more likely. Among the archaeozoological remains from the cultural layer of the Stare gmajne site, bone remains of a pig (*Sus* sp.) are quite common, but distinguishing between wild and domestic is usually difficult. At this point, we can only assume that pigs coprolites would not occur together with humans and neither in/along the vessels.

<sup>28</sup> e.g. TOLAR, T. et al. 2011.

<sup>29</sup> BOUCHET, F. et al. 2003; GONÇALVES, M. L. C. et al. 2003.

<sup>30</sup> WILKINSON, K., STEVENS, C. 2003; JACOMET, S. 2007.

<sup>31</sup> KUBIAK-MARTENS, L. 2022b, pp. 71-72.

<sup>32</sup> MAIER, U. 2001.



## Coprolite C2

Coprolite C2 was found during excavation and was stored in distilled water until multi-proxy analyses were performed. Macro-remain analyses show lots of chewed bone fragments, not only of fishes, but also of birds and/or mammals, and very little plant macro-remains (acorn and white goosefoot fruit fragments), what indicate canid (dog)-like origin of the coprolite C2. The assumption could be supported also with significantly less *Trichuris* sp. eggs (only 1; Tab. 3). The absence of other parasites, especially tapeworms (e.g. *Diphyllobothrium/Spirometra*) which were found in the coprolites from 2007 excavations (i.e. SG1 and SG5; see Tab. 4) can not be explained at the moment.

The relatively wide spectra of plants, represented by pollen in C2, suggests spring to early summer period flowering of i.e. *Corylus*, *Alnus*, *Iris*, *Betula*, *Quercus*, *Picea*, *Tilia*, etc.

## Comparison to the firstly analysed coprolites from Stare gmajne 2007 (trench 3)<sup>33</sup>

Table 5: Three coprolites from Stare gmajne 2007 (trench 3) that were macro- and/or micro-analysed.

	SG1	SG5	SG6
ARZOO	fish cranial elements, teeth; mammal, birds bones	fish vertebrae, cranial el.	n.d.
ARBOT	<i>Chenopodium</i>	no seeds	n.d.
PALYN	Cyperaceae, <i>Corylus</i> , <i>Quercus</i> , <i>Fagus</i> , <i>Alnus</i> , <i>Ranunculus</i> , Cerealialia (2)	n.d.	n.d.
PALPAR	<i>Diphyllobothrium/Spirometra</i> (8)	<i>Trichuris</i> (2), <i>Alaria</i> (11), <i>Diphyllobothrium/Spirometra</i> (4)	<i>Trichuris</i> (1)

n.d. – not done, no data

Scarce seed content and the absence of cereals (except of pollen) and other possibly nutritional plant taxa in all six investigated coprolites from Stare gmajne, trench 3 (2007),<sup>34</sup> besides prevalent fish cranial elements with teeth and other animal bones (i.e. birds, mammals in three out of four macro-investigated coprolites), testify that we were dealing with the dog rather than the human coprolites.

The result is partly confirmed with palaeoparasitological analysis, which shows the presence mainly of tapeworms (*Diphyllobothrium/Spirometra*) and flukes (*Alaria*) in two of the analysed coprolites. Such parasites transmit to mammals through consumption of raw or undercooked fishes (especially in the case of *Diphyllobothrium*), for *Spirometra* through the meat of various animals. The recovery of *Diphyllobothrium* in SG1 and SG5 is linked to the consumption of undercooked, smoked or raw fish by a fish-eating animal (carnivores or omnivores). Humans can also be infested. Many fish species can act as intermediate host in the parasite life-cycle. The genus *Alaria* in SG5 is particularly present in wild or domestic canids after the ingestion of frogs or toads. Eggs of whipworms, i.e. genus *Trichuris* were identified in two coprolites (SG5, SG6), but in smaller numbers. Infestation with whipworm occurs when the host ingests food or water polluted with eggs. Retrieved parasites, particularly the presence of *Alaria* sp. in SG5, seem to confirm a canid (i.e. dog) origin of the coprolites.<sup>35</sup>

During the analysis of the second series of samples (C1, C2, S3; see Tab. 3), *Echinostoma*, a new parasitic taxon could be identified in C1 coprolite. Unfortunately, this does not provide more information on the mode of contamination, due to the impossibility of attributing it to a specific species. At least 16 species of *Echinostoma* can infest human.<sup>36</sup> Concerning the other taxa, i.e. *Trichuris* and *Diphyllobothrium*, they could both infest humans or canids. The average sizes of *Trichuris* eggs (55 x 25 µm) are consistent with the human/pig host. Considering all these elements, the parasitic assemblages highlighted within new samples (excavated in 2021) tend towards that C1 could be of human/pig biological origin, which is not consistent to C2 and to

<sup>33</sup> see TOLAR, T. et al. 2021.

<sup>34</sup> See also other analyses (in total 6 coprolites were analysed) in: TOLAR, T. et al. 2021.

<sup>35</sup> TOLAR, T. et al. 2021, pp. 111-112.

<sup>36</sup> FELDMAN, M. et al. 2006.

previous coprolites analyses (i.e. of SG1, SG5, SG6; see Tab. 4). The sediment sample taken just below the C2 (i.e. from the cultural layer) is interestingly more similar to C1 than to C2, concerning parasite spectra.

Concerning palynological analyses of coprolites SG1 from 2007 and C2 from 2021, both have lots of similarities, but also some differences. In both samples, Cyperaceae pollen grains are the most dominant, which shows that both exterminators lived in a rather humid area. The SG1 sample was largely dominated by arboreal taxa (i.e. *Fagus*, *Quercus*, *Corylus*). A rather high proportion of cereals in C2 is surprising. This could be the consequence of both, the period when the defecation occurred (i.e. the time of cereal flowering) and/or of what the animal/person has drunk and/or eaten. Both SG1 and C2 have high percentages of pollen of early successional taxa, indicating early spring (SG 1) and spring to early summer (C2) deposit period.

Humans, pigs and the dogs were established as the dominant producers of the confirmed coprolites also e.g. in the Dutch project “*Neolithic Human Diet Based on Studeis of Coprolites ...*” which aim was to assess the diet and the health of the Late Neolithic populations.<sup>37</sup> According to the GS-MS assessment of the initial lipid extractions, there were some traces of possible acid compounds in several samples, suggesting a human or likely-human origin for 8 coprolites and an animal origin for three (including 2 pigs). The concentrations of lipids in 14 coprolites were too low to enable the determination of the source organisms.<sup>38</sup> The dominant presence of fish remains in all of the 16 analysed coprolites from the Dutch sites indicate that fish were frequently eaten, perhaps on a daily basis. Fish bone remains included vertebrae, fish scales and head bones (including teeth) of a very small cyprind and pike fishes, suggesting that the whole fishes were eaten, also by humans. They were probably cooked in pots, which would have softened the bones. Cereals (epidermis, phytoliths, pollen) were detected in 5 coprolites.<sup>39</sup>

## CONCLUSION

The initial question “*Dog or its master*” proved to be on spot. The results of the new analyses on new coprolites (i.e. C1, C2) from 2021 (trench 4) excavation confirm to be of different origin from those of the previous ones (SG1, SG5, SG6) in 2007 (trench 3) excavation. The parasitic spectra complement the data of macro-remains. A mixture of human/pig and canine coprolites could be highlighted at Stare gmajne site, which demonstrates a certain co-habitation, with a common waste management. Pollen analyses reveal an environmental vegetation with mostly hygrophilous species. Pollen found in coprolites could have reached the human/animal intestines by inhalation or in food/drinks. All palaeoparasitologically analysed coprolites and sediment sample contained eggs of intestinal parasites. Infection occurs by ingesting mature eggs in polluted soils, water or food, and is related to bad body hygiene and inappropriate waste management, which is often discovered at archaeological sites.

The presence of *Trichuris* and other parasites may indicate the fouling of the soil with faeces, which implies the long-term presence of humans (and animals) at one site. We are probably dealing with a mixed coprolite assemblage, which means that our contribution may be regarded as the reconstruction of the community diet at Stare gmajne site, where people were living together with their dogs (and perhaps even pigs) nearby, and likely all shared a similar diet. The hygienic conditions at the settlement must have been poor, since both humans and their animals were affected by multiple intestinal parasites.

Human/animal faecal remains are often difficult to identify with certainty. The method which proved to be quite useful is gas chromatography-mass spectrometry (GC-MS), where faecal lipid biomarker analysis is applied to confirm and identify the faecal origin.

Preliminary analyses from Slovenia indicate that we are dealing mostly with dog coprolites, particularly those containing bone fragments not derived from fish, while Kubiak-Martens et al. suggest that prehistoric human diet mostly based on whole-bodied small fish. A coprolite from Stare gmajne (C1) is probably not from a dog, but most likely from a human (possibly also of pig). The next step would certainly be biochemical analyses, in-

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<sup>37</sup> KUBIAK-MARTENS, L., VAN DER LINDEN, M. 2022.

<sup>38</sup> KUBIAK-MARTENS, L. et al. 2022, p. 127.

<sup>39</sup> KUBIAK-MARTENS, L., VAN DER LINDEN, M. 2022.

cluding GC-MS evaluation of faecal steroids. Macroscopically identified plant and animal taxa are often used for harder, less digestible plant and animal parts. They could provide food plant use along with the evidence of cooking and processing methods. In the future, DNA analyses would also be important because genetically identified taxa are often utilized for softer, more digestible, especially plant parts that would indicate dietary and potential medicinal uses of plants. Macroremains, GC-MS and DNA are complementary methods and should not be considered substitutes for each other.

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# PES ALI NJEGOV GOSPODAR? INTERDISCIPLINARNA ŠTUDIJA KOPROLITOV S KOLIŠČA STARE GMAJNE

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## IZVLEČEK

Začetne raziskave sedmih poznoneolitskih pasjih koprolitov s kolišč na Ljubljanskem barju, ki so bili odkriti v letih 2007 in 2014, rezultati pa objavljeni v letih 2019 in 2021, so zdaj podprte z novimi raziskavami dveh na novo odkritih koprolitov iz leta 2021. Pelodne analize, rastlinski in živalski makroostanki ter ohranjeni paraziti v njih kažejo, da smo tokrat naleteli ne samo na pasje, pač pa morda tudi na človeške iztrebke, torej na dva različna izvora. V članku so predstavljene razlike.

## KLJUČNE BESEDE

koproliti, pes, človek, eneolitik, kolišča, Ljubljansko barje, Slovenija

## POVZETEK

Začetne raziskave dobro ohranjenih, z vodo prepojenih več kot 5000 let starih koprolitov s kolišč na Ljubljanskem barju so pokazale, da gre za pasje iztrebke. Ugotovili smo, da predstavljajo tovrstne najdbe izjemno potencial za pridobivanje dodatnih informacij o življenju na koliščih, npr. status psa za človeka, prehranske navade, okolje, zdravstveno stanje, letni čas oziroma sezona iztrebljanja idr. Poleg potenciala in izjemno dobre ohranjenosti je bila povod za nova izkopavanja leta 2021 tudi sama številčnost koprolitov na tem najdišču. Leta 2007 je bilo v okviru 15 m<sup>2</sup> velike sonde namreč najdenih kar 16 pasjih koprolitov. Sondiranje v bližini sonde iz leta 2007 je bilo izvedeno leta 2021 v okviru bilateralnega raziskovalnega projekta JZ-2598. Odkrili smo novih devet pasjih ali človeških koprolitov, ki so bili podobnega velikostnega razreda in v bližini prvih, odkritih v letu 2007. Analizirana sta bila dva izmed njih (rastlinski makroostanki, ribji ostanki, pelod, paraziti, biokemizem). Trenutni rezultati kažejo, da smo tokrat naleteli tudi na človeške ali celo prašičje iztrebke, in ne le na pasje, saj sta se analizirana koprolita med seboj razlikovala tako po strukturi kot tudi po vsebini. C2 vsebuje pretežno ribje kosti, vključno z vretenci in zobmi, a le malo ribjih lusk in le 3 fragmente rastlinskih makroostankov, C1 pa, nasprotno, vsebuje več ribjih lusk, malo oziroma nič kosti ter več rastlinskih makroostankov, tudi žit. Analiza jajčec notranjih parazitov v njih dodatno prepozna koprolit C1 za bolj verjetno človeškega (lahko tudi prašičjega) kot koprolit C2. Zanimivo je tudi, da je bil C1 najden v sedimentu na dnu posode. Biokemijske analize še potekajo. Zaradi očitnega dvojnega izvora iztrebkov (pes, človek/praišč) smo se odločili analizirati še 5 od skupaj 9 koprolitov, najdenih v letu 2021, ki so se večinoma nahajali na le dveh kvadrantih (tj. 2 m<sup>2</sup>). Nekaterne analize so končane, na druge še čakamo. Ugotavljamo tudi, da so iztrebki (kot tudi ostanki v njih), najdeni v letu 2021, slabše ohranjeni kot tisti, ki so bili najdeni pred tem. Najdišče Stare gmajne je eno najbolj ogroženih še ohranjenih kolišč na Ljubljanskem barju, saj na njem poleg izsuševanja vsakodnevno poteka poljedelska mehanska aktivnost, ki neposredno vpliva na arheološke ostaline, ohranjenosti v ilovnatih plasteh le nekaj deset centimetrov pod površjem.