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Characterising life in settlements and structures: Incorporating faecal lipid biomarkers within a multiproxy case study of a wetland village

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ABSTRACT

Roundhouses are ubiquitous features of Iron Age landscapes across North West Europe, yet the way they were used internally is not well understood. We demonstrate how spatial analyses of steroid lipid biomarkers advances our understanding of household activities, living conditions and animal management associated with a well-preserved 5th century BCE roundhouse from Scotland's first Iron Age wetland village, Black Loch of Myrton, especially when combined with more traditional archaeological approaches. Faecal steroids (5β-stanols and bile acids) are well preserved within the wetland roundhouse floor deposits. Diffuse faecal inputs are identified within these deposits, limiting the resolution of faecal source discrimination compared with studies of concentrated faecal remains. However, analysis of both 5β-stanols and bile acids enables discrimination between ruminant (sheep, goat and cattle), pig and horse/human faecal remains. By integrating faunal data and entomological dung indicators we are able to characterise the on-site presence of animals associated with these archaeological structures. Steroids indicate short-lived and/or temporary pulses of dung deposition within the Iron Age roundhouse case study structure, which can be very difficult to determine using other archaeological proxies. Furthermore, our multiproxy results demonstrate the molecular preservation of steroids within deposits that have been subjected to regular floor cleaning, which is associated with the removal macrofossil proxies. Comparisons of multiproxy faecal signatures of the inner and outer sections of the structure show temporal and spatial heterogeneity in usage and living conditions. The faecal signature points to temporary sheltering of animals within the inner section of the structure. The multi-use and division of different activities within the roundhouse, determined by steroids, marks an important contribution to broader archaeological debates surrounding structures, their functions and re-use.

1. Introduction

A key advantage of analysing occupation sedimentary deposits, such as floor remains, is the retention of a wealth of information about the use of space in settlement sites (e.g. Manzanilla and Barba, 1990; Middleton and Price, 1996). The characteristics of these structural space uses, which may vary over time, can provide insights into social statuses and roles of houses, animal husbandry practises, food storage, and handcrafts etc. although, as is the case of Alpine Neolithic settlement houses, special functions are rare (Ebersbach, 2013). Almost all environmental proxies have been trialed to reconstruct the use of internal space including geochemistry, molecular proxies, pollen, insects, phytoliths as

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well as the standard analysis of micromorphology, plant macrofossils and faunal remains. The most effective characterisations rely on a combination of these proxies to provide multiple lines of evidence to support interpretations (Shillito, 2017). However, integration of multiproxy analyses can be complex and should be considered at the project design stage (Shillito, 2017) with clear considerations for the specificity of results obtained from each proxy (e.g. Middleton et al., 2010) as well as the role of the depositional environment as a record of activity (Shahack-Gross, 2011).

Of the biological proxies used to characterise occupation deposits, insects have been widely used due to their early synanthropism (Smith et al., 2020), and host-specific diversity related to almost all aspects of within structure activities as well as the external environment. Some of the best-known examples include the study of Norse North Atlantic farmsteads (Panagiotakopulu et al., 2007) and Viking age houses from 9th AD century Dublin (Reilly et al., 2016). Whilst pollen is less commonly used to characterise occupation deposits than insects, the case study of Pueblo houses in southwest USA demonstrates the ability of pollen spectra obtained from floors to suggest different room uses such as food processing, ceremonial function or meeting rooms (Morris, 1986). A non-in situ example includes the high concentrations of cereal and grazing indicator pollen adjacent to *crannogs* (artificial island settlements) taken to indicate crop storage/processing (O'Brien et al., 2005) and animal tethering and slaughter (Brown et al., subm). The use of phytoliths is more common in dryland settlement sites such as Çatalhöyük in Turkey (Ryan, 2011; Shillito and Ryan, 2013), but they have been used successfully in temperate European environments such as Williamson's Moss in Britain (Wade et al., 2019) and have great potential in tropical wetland sites such as the Kuk swamp in Papua New Guinea (Golson et al., 2017).

A commonly applied technique to assess function and use of space is micromorphological analysis of floors with soil phosphate and multielemental analysis (Middleton, 2004). Elevated soil phosphate is associated with animal use in a wide variety of environments (Holliday and Gartner, 2007). However, its equifinality has been demonstrated (Middleton et al., 2010) and accumulation patterns within soil requires careful interpretation (e.g. Nielsen and Kristiansen, 2014), particularly in wetland contexts that are impacted by changes in solubility, absorption, resorption, mobilization and leaching, at low pH and Eh, of sediments. In the classic Butser House, England, experiment, Macphail et al. (2004) showed how crust formation was important for phosphate retention and microscopic crust formation, the degree of floor compaction and its mineral content all related to the variability in phosphate depletion from floor surfaces exposed to pedestrian traffic and house cleaning. At the same experimental site, Evershed et al. (1997) highlighted that a manured area could not be clearly identified using concentrations of total phosphorus, but it was detectable using faecal lipid biomarkers (5β-stanols).

Recent developments in the refinement of faecal lipid biomarker signatures (Prost et al., 2017; Harrault et al., 2019) now facilitate the application of this approach within widely available bulk anthropic sediments, as well as concentrated faecal remains, to characterise animal husbandry and living conditions. These lipid compounds - termed steroids - have the ability to enhance characterisations of activity areas since they are direct markers of faecal matter produced by higher vertebrates and can identify human-animal interactions. The steroid composition of faecal matter produced by different animals varies according to their food sources, digestive processes and gut bacteria (Leeming et al., 1996). Therefore, diagnostic ratios of faecal and non-faecal sterols (e.g. 5β-stanols vs 5α-stanols) and bile acids can discriminate between human, porcine and herbivore faecal matter (Bull et al., 2002; Prost et al., 2017; Harrault et al., 2019). Steroids have identified the presence and source of faeces in a range of archaeological settings including coprolites and manure (Evershed et al., 1997; Bull et al., 2001; Shillito et al., 2011; Prost et al., 2017; Ledger et al., 2019) and archaeological soils (Simpson et al., 1998; Bull et al., 1999; Harrault et al., 2019).

The utility of incorporating steroids within studies of activity areas has been demonstrated using sterol analyses of deposits obtained from experimental settlements and palaeosols. For example, the combined analyses of elements and sterols from an experimental Iron Age settlement identified separate activity areas and provided positive identification of activities in all except one area (Hjulström and Isaksson, 2009). The first spatial analysis of sterols obtained from paleosols identified patterns of animal husbandry from land adjacent to a *ca.* 5th-11th c. AD Russian "fortress-settlement" (Harrault et al., 2019; Anderson et al., 2019). Whilst these studies showcase the ability of sterols to identify spatial patterns of activity areas and animal husbandry, the use of both sterol and bile acid analyses within wetland archaeological settlement sediment deposits has yet to be tested.

We present the first multiproxy spatial study of Iron Age roundhouse wetland sedimentary deposits from the Black Loch of Myrton (BLM) in southwest Scotland, UK (Fig. 1; Crone et al., 2018) using steroid lipid biomarkers (sterols and bile acids), ecofact analysis and micromorphology to investigate the use of space within a roundhouse structure (Structure 2). Excavations of waterlogged Iron Age roundhouses are rare, but other examples from the UK include Flag Fen (Pryor, 2001) and Glastonbury Lake Village (Hill et al., 2018). The BLM excavation offered an opportunity to investigate the usage of an Iron Age roundhouse since the nature of the wetland site means there was excellent structural integrity, providing insight into structural form and construction of the roundhouse (Crone et al., 2018), as well as good organic matter preservation within the archaeological soils. Structure 2 has well-stratified organic rich matrix, important for faecal steroids, which have low water solubility and are absorbed to particulate organic matter preventing vertical movement via leaching (Lloyd et al., 2012). As a result, steroids remain in situ at the point of deposition (Lloyd et al., 2012), are likely well preserved over the Iron Age timescale (Lin et al., 1978; Bull et al., 2001; Prost et al., 2017) and, in the case of coprolitic sources, are preserved in wetland settings (Ledger et al., 2019).

Two models for Iron Age roundhouse space use exist: (1) inner sections are areas of active communal domestic activity, with the outer section as a peripheral area for sleeping and storage (Hingley, 1990); and (2) outer sections of the roundhouse are reserved for stalling of animals (e.g. Kelly, 1988; Banks, 1995). The difference between these two models is dependent on region (Hill, 1995), with centrally focused roundhouse activity areas highlighted in the first model, generally found in northern regions of Iron Age Britain. To determine the most appropriate model of roundhouse use and to establish whether livestock co-habited spaces with people we need to establish what these inner and outer spaces were used for by integrating multiproxy indicators of humans and animals.

2. Methods

2.1. Study site

The Black Loch of Myrton (BLM) is a drained wetland in southwest Scotland, UK (54°45′13″N 4°32′53″W; Fig. 1). Recent excavations show the settlement was constructed on top of a natural peaty island approximately 50 × 60 m within a shallow fen marshland (Crone and Cavers, 2015, 2016). Excavations and dating (radiocarbon-dating and dendrochronology) of five of the settlement mounds show that the date of settlement at BLM was the latter half of the 5th century BCE, ending in the 3rd century BCE with at least three phases of construction and renewal (Crone et al., 2018).

Structure 2 is a large roundhouse 12.8 m in diameter (Fig. 1; Crone et al., 2018), the inner and outer sections of which were divided by a ring of posts proximal to the central stone hearth (Crone et al., 2018), likely reflecting a common, conscious organising principle of Iron Age roundhouse structures in Britain (Pope, 2007). The stratigraphy of the hearth, entrance and floor deposits indicate they have been refurbished

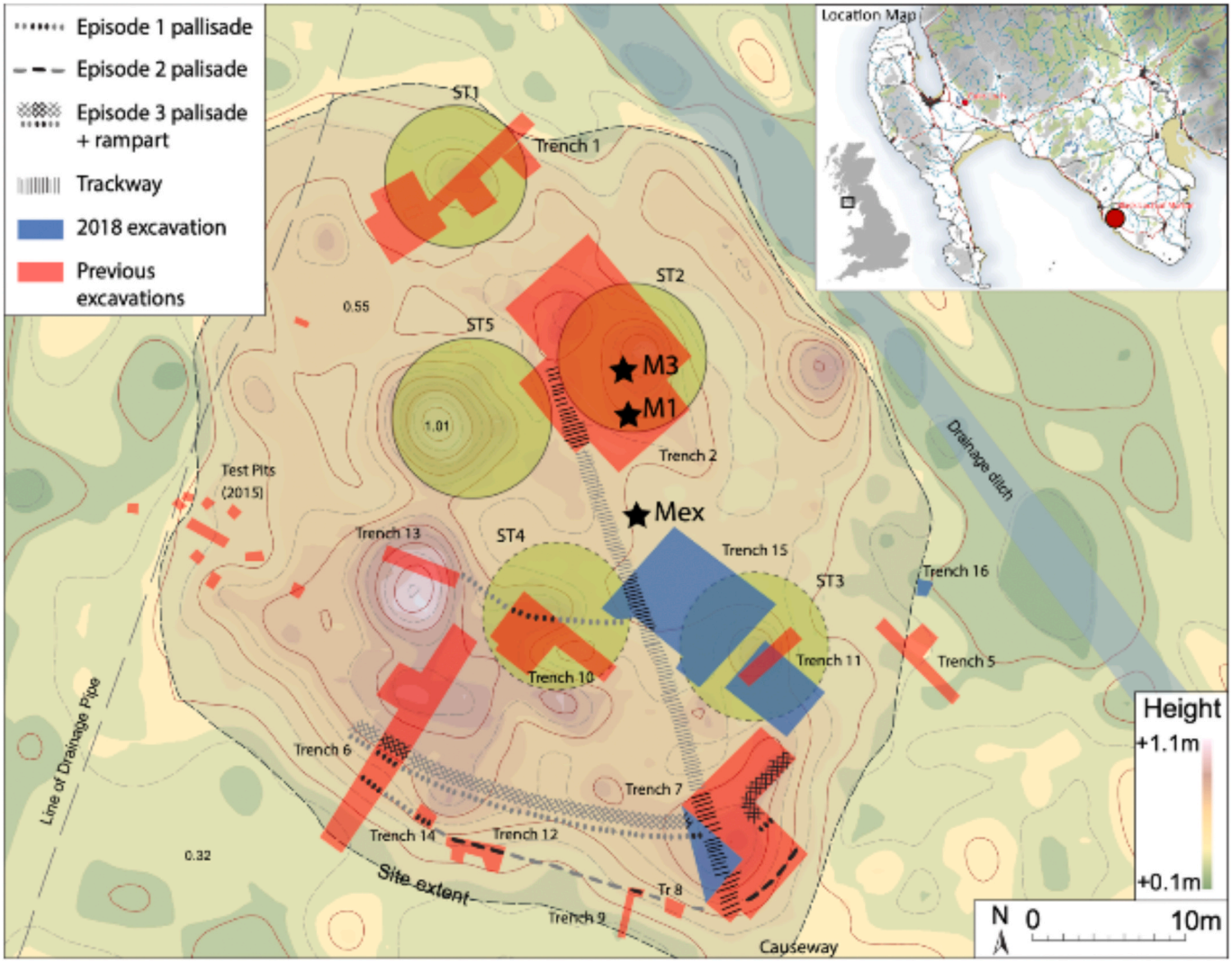

Fig. 1. Location of Black Loch of Myrton in southwest Scotland. Digital terrain modelling characterizes the topography of the site, revealing seven-eight discrete mounds, five of which have been excavated. Black stars indicate Structure 2 (ST2) sampling locations: M3 = inner roundhouse, M1 = outer roundhouse, M_{ex} = outside roundhouse entrance.

at least twice, leading to the build-up of stratified acidic layers of plant litter (pH 5.3 ± 0.4), which were used to create the floor surfaces (Crone et al., 2018). Chronological evidence for the construction, occupation and abandonment of Structure 2 brackets it to a 30 to 40-year period from *ca.* 435 BCE – 400 BCE (Crone et al., 2018). Preservation of structural and organic material is excellent due to waterlogging. Despite high-levels of organic matter preservation in BLM Structure 2, evidence for activities that took place within the roundhouse are limited: minimal material culture was recovered (Crone et al., 2018) and the micromorphology and macrofossil remains suggest regular cleaning within the structure, thereby removing anthropogenic activity signals (Robertson and Roy, 2019).

2.2. Sampling

Monolith tin samples were taken in summer 2015 from the inner and outer area of Structure 2 (Fig. 2). An additional monolith for organic geochemical analysis was obtained *ca.* 5 m outside of the structure from contemporary archaeological deposits in front of the roundhouse entrance in January 2017, to characterise external dung deposits and/or trampled dung originating from animals entering and leaving the structure (M_{ex}; Fig. 2). Samples for steroid analysis and micromorphology were extracted from the internal monoliths at depths corresponding to assigned contextual changes consisting of foundation deposits, primary floor layers and subfloor layers (Crone and Cavers, 2015, 2016).

2.3. Faecal steroid analysis

Total lipids were extracted from approximately 1 g of dried, homogenised sediment, spiked with internal standards (androstanol and hyocholic acid), with solvents (DCM:MeOH, 2:1, *v/v*) using microwave assisted extraction (heated to 70 °C over 10 min then held at 70 °C for 10 min; Kornilova and Rosell-Melé, 2003) and saponified using 5 M sodium hydroxide in MeOH. Following Bull et al. (2001), extracts were separated into neutral and acid fractions using aminopropyl SPE columns and these fractions were further split using silica gel column chromatography to isolate the sterol fraction and, following methylation using trimethylsilyldiazomethane (TMS-DAM) in toluene/methanol (4:1 *v/v*), the hydroxylated carboxylic acids (containing bile acids). The sterol and bile acid fractions were trimethylsilylated using *N,O*-bis(trimethylsilyl)trifluoroacetamide (BSTFA)+ trimethylcholorosilane (TMCS) (99:1 *v/v*).

Both derivatized sterol and bile acid fractions were dissolved in

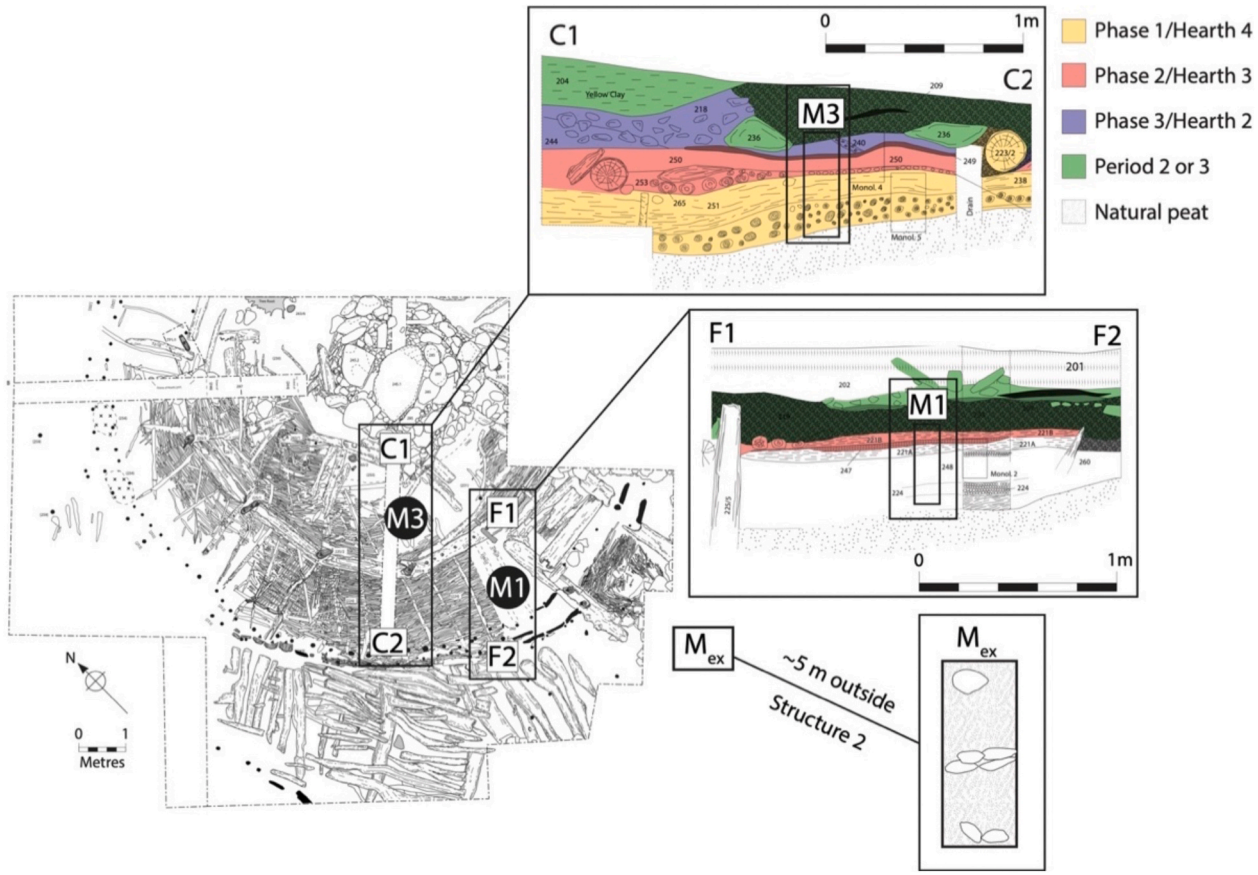

Fig. 2. Location of monolith samples obtained inside (M1, M3) and outside (M_{ex}) Structure 2.

50–100 μL of ethyl acetate prior to analysis by gas chromatography-flame ionisation detection (GC-FID) and gas chromatography-mass spectrometry (GC-MS). GC-MS analyses were performed using a ThermoScientific ISQ, with an ion source temperature of 300 °C and electron energy of 70 eV. The analyser was set to scan *m/z* 50–650 with a duty cycle time of 0.2 s. Chromatographic separation was performed on an Agilent fused silica capillary column (HP-5, 60 m × 0.25 mm ID × 0.25 μm df). Sterol derivatives were analysed using the following temperature programme: 50 °C (held for 2 min) to 200 °C at 10 °C min^{-1} then to 300 °C at 4 °C min^{-1} and held for 20 min. Bile acid derivatives were analysed using the following temperature programme: 40 °C (held for 1 min) to 230 °C at 20 °C min^{-1} then to 300 °C at 2 °C min^{-1} and held for 20 min. GC-MS peaks were identified through comparisons with known mass spectra (NIST08; Prost et al., 2017 and a laboratory reference library), example chromatograms (Prost et al., 2017) and standards where possible. Analytes were quantified based on internal standards.

Potential faecal sources were identified from the sterol fraction using a ratio of the sum of faecally derived cholesterol reduction products (coprostanol + epicoprostanol) to the sum of environmentally and faecally derived cholesterol reduction products (5α-cholestanol + coprostanol + epicoprostanol) (Ratio 1; Bull et al., 1999) with ratio values *ca.* ≥0.3 indicative of potential faecal matter input (Prost et al., 2017).

$$\frac{(coprostanol + epicoprostanol)}{(5\alpha - cholestanol + coprostanol + epicoprostanol)} \quad \text{(Ratio 1)}$$

Ratio 1 does not definitively identify faecal matter in isolation since small proportions of these compounds are also produced by the reduction of cholesterol in the natural environment (primarily to produce 5α-cholestanol), thereby requiring comparative controls. The identification of herbivore faecal matter was indicated by the C_{27} to C_{29} 5β-stanol ratio (Ratio 2; Leeming et al., 1997), with values < 0.38 indicative of herbivore faeces.

$$\frac{(coprostanol)}{(coprostanol + 5\beta - stigmastanol)} \quad \text{(Ratio 2)}$$

Evidence for the presence of faecal matter was also supported by the presence of bile acids and the dominant faecal matter source was identified using the ratio of deoxycholic acid (DCA) to lithocholic acid (LCA) ratio (Prost et al., 2017). Based on modern experimental data, the values of this ratio can be ascribed in the following way: <0.4 pigs and/or geese; 0.6–4.5 humans and/or horses; >5 ruminants (cattle, sheep and goats) (Prost et al., 2017). Whilst the dominant faecal source can be identified using these ratios, this does not preclude the presence of other faecal sources in smaller quantities.

2.4. Insect analysis

Six bulk sediment samples (2–5 L) from floor contexts were processed using the standard paraffin floatation protocol (Coope, 1986). Briefly, sediments were wet-sieved through nested sieves (3 mm and 300 μm) to remove the inorganic clay and silt fraction, respectively. The collected float was washed with detergent then rinsed and stored in ethanol. Insect remains were picked using a large Bogorov sorting tray under a stereo microscope (× 10 - × 60 magnification) and the insects placed in ethanol for storage.

Beetle remains were identified using modern reference collections and standard published keys (e.g. Lindroth, 1974; Foster et al., 2014) and recorded as Minimum Numbers of Individuals (MNI). The species list and associated ecological information were generated using BUGS-CEP (Buckland and Buckland, 2006), following the taxonomy of Duff (2008). Fly and ectoparasite remains were identified using reference materials and manuals (Skidmore, 1985; Smith, 1989; Whitaker, 2007);

lice and fleas were identified to species level when heads were available. Muscidae fly puparia were identified to species level whilst remaining individuals could only be identified to genus level. Results presented here are a subset of the insect assemblage data, which are published elsewhere (Davies et al., n.d.), focusing on taxa that display an exclusivity for foul environments, are very common in dung and are closely associated with animals, following Hall and Kenward (1990) and Smith (2012).

2.5. Animal and plant macrofossil remains

Bulk sediment samples were processed using the standard floatation

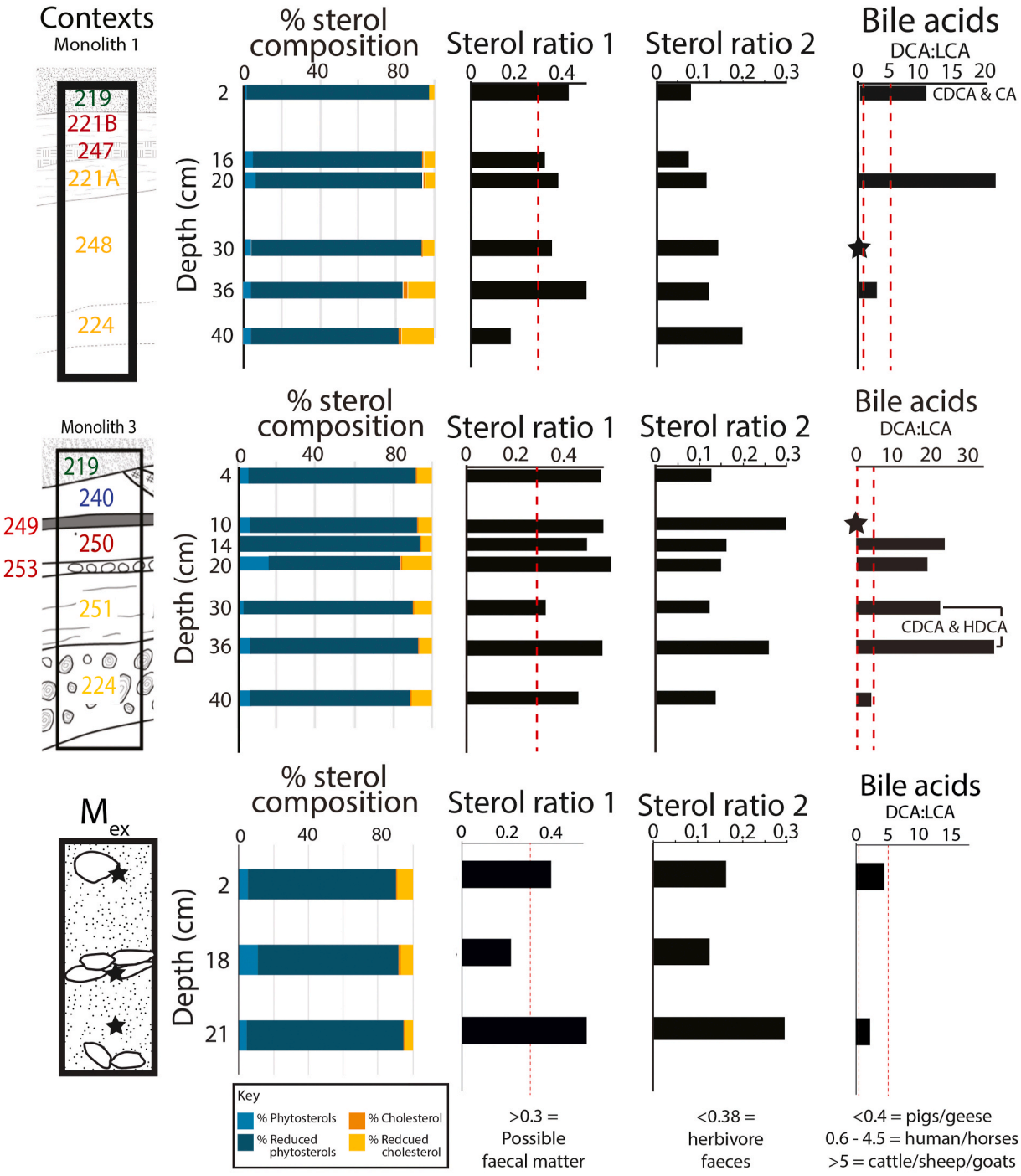

Fig. 3. M1 (outer), M3 (inner) and M_{ex} (outside roundhouse entrance) steroid characteristics. Contexts are coloured by assigned phases (Phase 1: yellow; Phase 2: red; Phase 3: blue; Period 2 or 3: green). The phytosterols (campesterol and sitosterol: light blue) and their reduction products (C_{28} and C_{29} stanols: dark blue) and those of cholesterol (C_{27} stanols: orange and yellow), reveal the degree of biohydrogenation of the Δ^5 unsaturated sterols. Sterol ratios refer to those numbered in the main text: (1) is indicative of the possible presence of faecal matter when >0.3 and (2) is indicative of herbivore faecal input when <0.38. Bile acid DCA:LCA ratios indicate dominant faecal source <0.4 pigs/geese, 0.6–4.5 human/horses, >5 ruminants. Star symbols highlight where DCA was present in isolation, therefore whilst the diagnostic source value cannot be calculated, faecal matter is present within the sample. Samples containing chenodeoxycholic acid (CDCA), cholic acid (CA) and hyodeoxycholic acid (HDCA) are also indicated using bile acid abbreviations. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

method (Kenward et al., 1980), with waterlogged samples processed by hand to maximise recovery of fragile plant remains. Macrofossils were examined under a microscope (× 10 - × 100 magnification) and identifications were made using modern reference material and seed atlases (Cappers et al., 2006; Jacomet, 2006). Charcoal samples containing two or more wood species were designated as fuel waste, whilst those containing larger concentrations of a single species were interpreted as burning events. Bone was identified to element and species with the aid of reference material and skeletal atlases (Schmid, 1972; Hillson, 1986). Where an element could not be identified to species level, it was categorised into large mammal (cattle/horse/deer), medium mammal (sheep/goat/pig) and small mammal (dog/cat/rodent).

2.6. Soil micromorphology

Eleven samples were extracted from the internal Structure 2 monoliths, corresponding to contexts targeted for steroid analysis, and prepared for micromorphological analysis after Murphy (1986). Thin section description was conducted using the identification and quantification criteria by Bullock et al. (1985) and Stoops (2003). Abundance of fabric constituents were estimated following categories outlined by Stoops (2003). Deposit types were identified based on particle size, shape and the composition of the coarse and fine fraction, particularly the frequency and type of organic matter, minerals and anthropogenic inclusions. Trampling was indicated by linear and parallel distributions, polyconcave voids and platy microstructures (Courty et al., 1989; Milek, 2012).

3. Results

3.1. Faecal steroids

The total sterol composition of all samples is dominated by plant-derived compounds (phytosterols; campesterol and sitosterol and reduction products of phytosterols; C_{28} and C_{29} stanols), with cholesterol and its reduction products (C_{27} stanols) accounting for <10% (Fig. 3).

Ratio 1 returns values of >0.3, indicating a potential faecal input in all samples apart from two: [224] in M1 and 18 cm in M_{ex} (Fig. 3, Table 1 and 2). The lack of faecal matter in these two samples is confirmed by the absence of detectable bile acids. Ratio 1 of one sample ([219; M3]) suggests the possible presence of faecal matter but this is not supported by bile acids. Ratios of detected bile acids (DCA:LCA) constrain dominant animal faecal matter sources in Structure 2 deposits to pigs (<0.4), ruminants (cattle and/or sheep and/or goats (0.6–4.5) and human and/or horses (>5) (Fig. 3, Table 1 and 2). Ratio 2 < 0.38, indicates herbivore faecal input in all samples.

3.1.1. Spatial patterns in faecal steroids

Based on the DCA:LCA bile acid ratio (Prost et al., 2017) and sterol ratio 2 (Leeming et al., 1997), the dominant source of faecal matter within the roundhouse originates from ruminants (cattle, sheep and/or goats), with ruminant signals occurring more frequently in the inner section of the roundhouse (M3) than the outer section (M1) (Fig. 3, Table 3). Faecal matter from pigs and humans and/or horse are identified in some samples from both the inner and outer sections of Structure 2 based on the presence of hyodeoxycholic acid (HDCA), diagnostic of pig faeces, and chenodeoxycholic acid (CDCA), diagnostic of human and/or horse faeces (Prost et al., 2017). While evidence for human and/or horse faeces is detected in M_{ex}, outside of the roundhouse, there is no clear evidence of ruminant or pig faeces in this outside area (Fig. 3, Table 1).

Table 1
Summary of faecal steroid results from outside Structure 2 (M_{ex}) by depth. *Dominant faecal origin is based on bile acid profiles. All samples have sterol ratio 2 values indicative of herbivore faecal matter, therefore a mixed faecal deposit is likely.

Depth	Description	Steroid characteristics	Faecal origin*
2 cm	Organic rich deposit with large stones	Faecal steroids present	Humans and/or horses
18 cm	Organic rich deposit with stones	No faecal steroids present	None
21 cm	Organic rich deposit	Faecal steroids present	Humans and/or horses

3.1.2. Temporal patterns in steroid biomarkers

The first detection of faecal material occurs during Phase 1, registering earlier in the inner section of the structure than the outer section (Fig. 3, Table 1). The initial bile acid ratios in both the inner and outer sections of the roundhouse originate from humans and/or horses, then becomes mixed with input from humans and/or horses (CDCA), pigs (HDCA) and ruminant input (DCA:LCA ratio). The DCA:LCA ratio in Phase 2 indicates ruminant faecal matter and this is the only faecal source detected during this phase in M3 (inner). However, the source of faecal matter in the outer structure (in M1) switches to a human and/or horse dominated signal in latter contexts of Phase 2.

3.2. Ecofacts

3.2.1. Insects

Insect preservation is high with intact remains, although overall abundance is variable, with abundances ranging from 6 to 91 MNI L^{-1} of sediment (Table 3). The highest concentrations of beetles and ectoparasites are present within [221A] (Fig. 4) from inner Phase 1, but concentrations between samples from the inner and outer sections of Structure 2 are comparable. There are low abundances of beetle taxa in the 'foul rotting' category e.g. *Aphodius* spp. and *Aphodius distinctus* (Müll) (Atty, 1983), but they are present in samples from both the inner and outer sections in conjunction with species associated with the dung of large herbivores (e.g. *Cercyon quisquilius* (L.), *Aphodius prodromus/-sphacelatus* (Panz)/(Brahm), *Cercyon melanocephalus* (L.), *Aphodius contaminatus* (Hbst.)(Koch, 1989; Duff, 1993)). Lice were found in both phases, *Bovicola bovis* (cattle louse) and *Pulex iritans* (human flea) [250] (Phase 2, inner section) and *Bovicola ovis* (sheep louse) (Phase 1, outer section). Fly puparia are common in both Phases 1 and 2 but are more abundant in samples from the outer areas of the roundhouse.

3.2.2. Macroplant remains

Macroplant remains primarily consist of three categories: food and food processing waste; fuel debris and flooring materials (Table 4, Fig. 5). The food waste consisted of cereals and wild food sources with the majority of this material carbonised but with small amounts that are waterlogged. The cereal and wild food remains were detected within contexts from both the inner and outer sections of the roundhouse: cereal remains were concentrated in inner Phase 1 [251] and outer Phase 3 [219], and wild food remains were most abundant in [251], from inner Phase 1, but were also present in [248] (outer Phase 1), [247] (outer Phase 2) and [250] (inner Phase 2). The main type of fuel used was wood, represented by a mixture of species, but there was also a small quantity of charred peat. There was no evidence for other types of fuel such as dung in any of the contexts under discussion. The materials used for flooring consisted primarily of bracken, sedges, rushes, woodrush and woody brash.

3.2.3. Faunal remains

The majority of faunal remains recovered from Structure 2 were small burnt bone fragments, 90% of which were not identifiable to species. There were small quantities of unburnt bone and teeth present. The number of identifiable specimens present in Structure 2 were cattle (61), sheep/goat (4), pig (2), large mammals (47) and medium

Table 2
Summary of common dung and animal-associated insects from Structure 2 by location and phase (n.d. = non detected). MNI = Minimum number of individuals per L^{-1} (all taxa). Foul decomposers = beetle species primarily associated with foul, rotting organic matter (often dung) as defined by Hall and Kenward (1990) & Smith (2012).

Context	Description	Location	Phase	Conc. (MNI l^{-1})	Dung and Foul matter beetles (total counts)	Ectoparasites and flies (total counts)
251	Plant litter subfloor	Inner	1	24	*Aphodius Distinctus* (1)	*Musca domestica* (3) *Stomoxys calcitranus* (3)
250	Plant litter subfloor	Inner	2	32	*Cercyon quisquilius* (1) *Aphodius prodromus/sphacelatus* (1)	*Bovicola bovis* (3) *Pulex iritans* (1) Phthiraptera indet. (10) *Musca domestica* (13) *Stomoxys calcitranus* (4)
249	Carbonised plant litter	Inner	2	9	n.d.	n.d.
248	Branchwood and brash	Outer	1	48	*Cercyon melanocephalus* (1) *Aphodius contaminatus* (1) *Aphodius* spp. (1)	*Bovicola ovis* (1) Phthiraptera indet. (3) *Musca domestica* (4) *Stomoxys calcitranus* (4)
221A	Plant litter subfloor	Outer	1	108	*Cercyon pygmaeus* (1) *Cercyon unipunctatus* (1) *Aphodius distinctus* (1)	Lice spp. indet. (1) *Musca domestica (37)* *Stomoxys calcitranus (10)*
221B	Plant litter subfloor	Outer	2	32	*Aphodius ater (1)* *Aphodius* spp. (1)	*Musca domestica (46)* *Stomoxys calcitranus (14)*

Table 3
Summary of faecal steroid results from within Structure 2 by location and phase. Descriptions of floor layers from Crone and Cavers (2015). *Dominant faecal origin is based on bile acid profiles. All samples have sterol ratio 2 values indicative of herbivore faecal matter, therefore a mixed faecal deposit is likely.

Context	Description	Location	Sample depth (cm)	Phase	Steroid characteristics	Faecal origin*
224	Plant litter subfloor	Inner (M3)	A1: 40 A2: 36	1	Faecal steroids present	A1: Humans A2: Mixed source: ruminants, pigs, humans and/or horses
251	Plant litter subfloor	Inner (M3)	30	1	Faecal steroids present	Mixed source: ruminants, pigs, humans and/or horses
253	Small branchwood	Inner (M3)	20	2	Faecal steroids present, reduced decay indicators	Ruminants
250	Plant litter subfloor	Inner (M3)	14 10	2	Faecal steroids present, reduced decay indicators	Ruminants
249	Carbonised plant litter	Inner (M3)	8	2	n/a (low organics)	None
240	Orange clay floor	Inner (M3)	4	3	No bile acids	n/a
224	Plant litter subfloor	Outer (M1)	40	1	No bile acids	n/a
248	Branchwood and brash	Outer (M1)	36 30	1	Faecal steroids present	Humans and/or horses
221A	Plant litter subfloor	Outer (M1)	20 16	1 & 2	Faecal steroids present	A1: Ruminants A2: Pigs
247	Grey clay, subfloor	Outer (M1)	12	2	n/a (low organics)	None
221B	Plant litter subfloor	Outer (M1)	5	2	n/a (low organics)	None
219	Peaty clay, decomposed floor	Outer (M1)	2	Period 2/3	Faecal steroids present	Mixed source: ruminants, pigs, humans and/or horses

mammals (164) (Table 5; Fig. 5).

3.3. Micromorphology

The primary constituent of the floor material is plant organic matter, which is exceptionally preserved throughout the samples (Fig. 6), but differences exist in the birefringence of organic matter with less degradation exhibited in samples from the outer area of the structure. Occasional thin excremental pedofeatures (<100 μm) caused by microfauna, indicative of limited bioturbation, are restricted to outer Phase 1 [248] and inner Phases 1 and 2 [250] and [251]. Anthropic indicators, based on micromorphology, are limited in most of the samples from the outer contexts as there are very few small wood and bark chips. Small amounts of coprolitic material, although not identified to species level, was observed in the inner section from context [251] (Phase 1) and possibility [240] (Phase 3) (Table 6).

The identification of trampling, a possible transfer mechanism of faecal material in organic sediments under waterlogged conditions is difficult to detect, as water and compression during burial causes swelling of sediment and masks trampling indicators. However, the presence of loam and distinct microstructures suggests some trampling in both the inner and outer sections of the roundhouse during Phase 1 (contexts [224], [251] and [248]) but only in the inner section during Phase 2 [250] (Fig. 6, Table 6).

4. Discussion

4.1. Occupation floor deposits: detection of faecal matter and source organisms using steroid biomarkers

The majority of samples analysed from both the inner and outer sections of Structure 2 contained evidence of faecal matter as supported by the presence of bile acids, which are deposited in the excreta of vertebrates (Haslewood, 1967; Hofmann and Hagey, 2008).

Fig. 4. Example of preservation level of beetle remains from context [221A]. A large number of elytra are from the hydrophilid genus *Cercyon* (especially *Cercyon analis*) alongside staphylinids and other hydrophilids.

Table 4
The waterlogged and carbonised macroplant assemblage for food and food processing waste and fuel debris categories. Key: *≤ 10, ** = 10–29, *** = 30–100, ****≥100. All carbonised macroplants are recorded in brackets and all other plant remains are preserved through waterlogging.

Phase		**1**	**1**	**1**	**2**	**2**	**2**	**2**	**3**	**3**
Location (Out = outer, In = inner)		**Out**	**Out**	**In**	**Out**	**In**	**In**	**In**	**Out**	**In**
Context		**221**	**248**	**251**	**247**	**249**	**250**	**253**	**219**	**240**
Sample Vol (kg)		2.5	2.5	2.5	2	1.9	2.5	0.7	20	5
% Sorted		50	50	50	50	50	50	100	100	100
Vernacular name	**Plant part**									
Hordeum vulgare L.	Caryopses								(*)	
Hordeum var *nudum* L.	Caryopses								(*)	
Hordeum sp.	Caryopses			*					(**)	
Triticum dicoccum L.	Caryopses								(*)	
Triticum dicoccum L.	Glumes			**					*	
Triticum dicoccum/spelta L.	Caryopses								(**)	
cf. *Triticum aestivum/compactum* L.	Caryopses								(*)	
Triticum sp.	Caryopses								(**)	
Triticum sp.	Glume		*	***						
Cerealia indet.	Caryopses		**	* (*)					* (*)	
Cerealia indet.	Glume									(*)
Wild food										
Corylus avellana L.	Nutshell frgs		*(*)	*			*			
Corylus avellana L.	Buds and/or bud-scales				*					
Rubus idaeus L.	Seeds			***	*(*)					
R. fruticosus agg	Seeds				*					
Fuel										
Charcoal (weight in g)			5.5	2.5		2.5		11.5	56.2	91
Charred peat			*	**		****	**			

Comparisons of bile acids and 5β-stanols within this study highlight the importance of considering context when applying sterol ratio threshold values (Grimalt et al., 1990; Bull et al., 1999) to definitively identify faecal sources within wetland settlement deposits: all ratio 1 values within this study were <0.7, which would only indicate the *possibility* of a faecal source based on Grimalt et al. (1990) thresholds, despite the majority of samples analysed within Structure 2 containing conclusive evidence of faecal matter deposition based on bile acids profiles. The ratio 1 threshold value was designed for modern sewage samples (Grimalt et al., 1990) and its validity in archaeological contexts has been critiqued (Bull et al., 1999, 2001; Bull et al., 2005; Simpson et al., 1998; Prost et al., 2017). The application of ratio thresholds to identify faecal inputs has been shown to be particularly challenging within organic rich soils, such as those obtained from Structure 2, owing to the abundance of 5α-stanols derived from plant remains (Birk et al., 2011); this drives faecal sterol ratio below the indicative thresholds even when faecal matter is present (e.g. Fitzsimons et al., 1995; Birk et al., 2011).

Whilst the dominance of a 5α-stanol input in organic rich soils could

Fig. 5. Examples of macroplant and faunal remains extracted from Structure 2 bulk samples. A: hazelnut [219], B: burnt bone [219], C: cereal grains [219], D: charcoal [219], E: chaff [240].

Table 5
Summary of burnt bone results from within Structure 2 by location and phase.

Context	Location	Phase	Bones present	Identifiable species/ mammals
251	Inner	1	1.6g, 7 fragments	Large mammal long bone shaft Medium mammal rib (cut mark)
253	Inner	2	0.01g, 2 fragments	None identifiable
250	Inner	2	None	
249	Inner	2	2.8 g, 14 fragments (8 unburnt)	Cattle bone Cattle premolar, molar Medium mammal rib
240	Inner	3	51.7g, 56 fragments (10 unburnt)	Large and medium mammal long bone shafts Medium mammal mandibles, rib and vertebrae
248	Outer	1	5 g, 12 fragments (6 partly charred)	Medium mammal long bone shaft x 2 (burnt), phalanx and partly charred premolar
221	Outer	1 and 2	None	
247	Outer	2	None	
219	Outer	Period 2/3	44.5 g, 26 fragments (<50 mm, burnt)	Cattle molar (unburnt) Sheep/goat humerus (burnt)

call for a lowering of faecal sterol threshold values, one sample analysed from the inner section of the roundhouse contained a sterol ratio within the 'possible faecal matter' range despite having no corresponding bile acids and therefore no supporting evidence for faecal matter. This example echoes findings from other studies reporting the presence of 5β-stanols despite no other evidence for faecal deposition (e.g. Bethel et al., 1994; Bull et al., 2001; Evershed et al., 1997). Multiple lines of faecal evidence, including both 5β-stanols and bile acids are therefore a more robust approach than changing threshold values when working with diffuse faecal sources in sedimentary settings.

Several studies have successfully circumvented sterol ratio threshold problems for faecal identification by comparing background sediment 5β-stanols concentrations with those from anthropic samples (e.g. Birk et al., 2011; Harrault et al., 2019). Alongside this contextual approach, our results demonstrate the importance of analysing both the sterol and bile acid lipid fractions from the same sediment sample when characterising diffuse faecal sources. This combined approach, also encouraged by Bull et al. (2002) and Prost et al. (2017) for more concentrated faecal inputs, not only provides greater confidence in faecal identification and constraining faecal sources, but also mitigates against possible difficulties in obtaining contemporary non-anthropic sediment samples required to accurately establish background concentrations. An example of such difficulty within the Iron Age setting of this study, is achieving adequate chronological control for comparisons of different sampling locations when dates fall within the Hallstatt plateau (Becker and Kromer, 1993), a period of minimal discernible changes in radiocarbon calibration curve between 750 and 400 cal BCE, when most radiocarbon

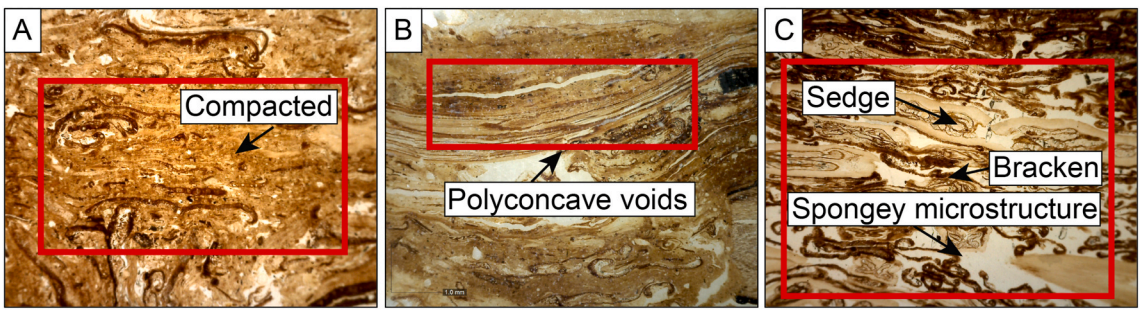

Fig. 6. Examples of morphology from Structure 2 samples. A: compacted/trampled organic layer [224], B: polyconcave voids [250] (inner), C: spongy microstructure and layers of bracken and sedge material [250] (outer).

Table 6
Summary of micromorphology results from within Structure 2 by location and phase (n.d = non detected).

Context	Location	Phase	Coprolitic material	Trampling indicators
224	Inner	1	n.d.	Lenticular microstructure. Loam/soil clasts brought in from outside embedded within matrix.
251	Inner	1	Unknown source	Compacted, lenticular to massive microstructure. Loam/soil clasts embedded within matrix.
253	Inner	2	n.d.	n.d.
250	Inner	2	n.d.	Linear compaction and striation of coarse material. Polyconcave voids
249	Inner	2	n.d.	n.d.
240	Inner	3	Possible herbivore	n.d.
224	Outer	1	n.d.	Parallel arrangement of inclusions, massive microstructures, polyconcave voids, low porosity, loam/soil clasts.
248	Outer	1	Soil microfauna only	Possible (dusty clay coatings to voids indicative of rotational movement of sediment caused by trampling)
221	Outer	1 and 2	Possible (yellow phosphatic filling)	n.d.
247	Outer	2	n.d.	n.d.
219	Outer	Period 2/3	n.d.	n.d.

determinations have calibrated age ranges in the order of several centuries (Crone, 2012).

Our steroid results demonstrate the presence of faecal matter from cattle/sheep/goats, pigs and horse and/or humans in Structure 2. The presence of ruminants is supported by the faunal evidence which includes bones and teeth from cattle and sheep/goat. There is no evidence for pig or horse faunal remains in the contexts analysed, although the presence of pigs on site is supported by faunal remains from other contexts and insect remains are indicative of large herbivores contemporaneous with human/horse faecal signals (Fig. 7). Steroid analyses of Structure 2 provide evidence for a greater diversity of animals associated with each context compared with faunal analysis where the acidity of the soils (pH 5.3 ± 0.4) hinders calcified bone survival.

The steroid results from Structure 2 have enhanced characterisation of animals associated with the roundhouse, however, the resolution of some identified faecal sources is lower than expected based on ratios obtained from modern reference material and concentrated archaeological faecal deposits (e.g. Prost et al., 2017; Harrault et al., 2019). This likely represents the difficulties of identifying faecal sources using diagnostic ratios and key indicator compounds when faecal inputs originate from a mix of source organisms (Prost et al., 2017) and are incorporated within organic-rich sedimentary archives (Birk et al., 2011). In such instances, the dominant faecal source organism(s) may be identified, but the refinement of faecal source identification relies on bile acid preservation (Prost et al., 2017). Faecal sterol distributions have also been used to refine differentiation of faecal sources, such as multivariate analyses of eleven 5β-stanol compounds (Harrault et al., 2019). However, the full suite of 5β-stanol compounds required for faecal source differentiation are not present in all lipid extracts, including samples from this study and those analysed by Leeming et al. (1996), thereby limiting the resolution of faecal source identification. Such differences in 5β-stanol characteristics between studies may relate to differences in diet, which controls compound distributions (Prost et al., 2017; Harrault et al., 2019), or the dominance of plant-derived sterols and other polar lipid compounds within organic-rich sediments, which mask low 5β-stanol concentrations despite extensive sample clean-up within the lipid analytical protocol.

Whilst multiple lines of steroid evidence must be considered when identifying faecal sources (Prost et al., 2017), an important consideration is the sensitivity of diagnostic ratios to different faecal sources. For example, since ruminants have a characteristic bile acid-derived DCA:LCA value, which is an order of magnitude higher than pigs and in some cases human and/or horses, their faecal signal has the potential to dominate a mixed source DCA:LCA ratio even if they were not the dominant faecal input. Therefore, whilst the dominance of ruminant faecal matter may be a robust feature within Structure 2 and is supported by sterol ratio 2, it may also be influenced by the sensitivity of the DCA:LCA ratio to ruminant faecal input. Experimental studies are essential to refine these diagnostic ratios and steroid distributions for diffuse faecal inputs within sedimentary deposits using approaches such as mixing models.

4.2. Multiproxy comparisons of faecal indicators

Approximately 60% of analysed floor deposits contained dung indicators within the steroids compared with 50% from insect analyses and 10–20% from micromorphology (Fig. 7). There are no conclusive dung indicators within the macroplant remains, although distinguishing between dung, fodder and floor deposits from macroplant remains is complex since plant assemblages are similar within these sources. Context [251] from Phase 1 of the inner section of the roundhouse does contain a high abundance of raspberry (*Rubus idaeus* L.) seeds, which may originate from faecal deposition (e.g. Buckland, 1976; Miller and Smart, 1984). Confirmation of faecal matter within this context is provided by the mixed steroid signal and the identification of coprolitic remains within the micromorphology (Fig. 7), thus demonstrating the value of multiproxy comparisons, as also presented by Shillito et al. (2011).

Multiproxy dung comparisons across Structure 2 demonstrate the presence of steroids, low abundances of dung/foul indicator insect species and minimal micromorphological and macrofossil evidence, which suggests dung deposits that are transient or restricted (rather than persistent or large scale) within the roundhouse. The low quantities of domestic debris and sharp contacts between floor layers point towards active floor cleaning and/or removal of dung from Structure 2 and may explain the low insect signal throughout the structure. Despite the removal of floor material, the geochemical faecal signature has been preserved within the remaining floor surfaces. Similar practices of floor cleaning have been identified at other Scottish Iron Age structures e.g. Cnip in Lewis (Armit, 2006) and Cults Loch in Wigtownshire (Roy, 2018; Robertson, 2018), where removal and replacement of floor layers were identified from excavated stratigraphy. Incorporating steroid analysis of archaeological structures therefore has the potential to provide a more holistic insight in to occupation conditions of Iron Age roundhouses. This is especially true where floor clearing has occurred and many of the more traditional microscopic anthropic signals have been removed, or preservation conditions for macro-organic materials is poor.

4.3. Multiproxy characterisation of Iron Age wetland roundhouse use

4.3.1. Spatial patterns of use associated with structure 2

Our steroid results show clear spatial differences between inside and outside of Structure 2, with ruminant bile acid profiles detected in M3 and M1, but are absent from outside in M_{ex} (Fig. 3). The ruminant faecal signal also differs within the structure, with a stronger ruminant signal present in the inner section of the roundhouse (Fig. 8). This faecal signal in the inner section is concomitant with *Bovicola bovis* (cattle louse) (Table 3, Fig. 7) and is consistent with evidence from the

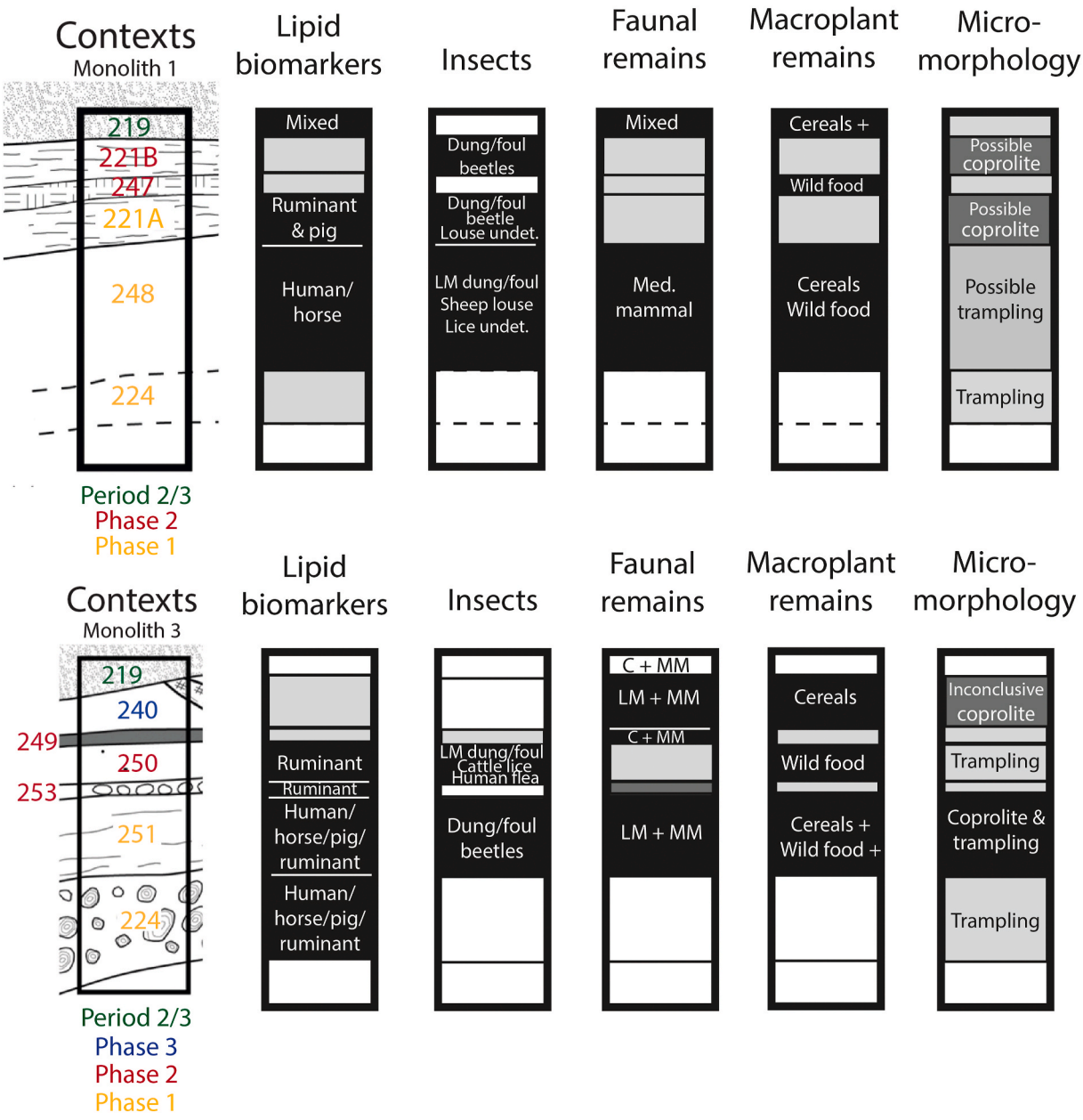

Fig. 7. M1 (outer) and M3 (inner) proxy comparisons. Black indicates clear evidence of large mammals, faecal sources or domestic food waste, dark grey indicates possible evidence of large mammals or faecal sources, light grey indicates no evidence of mammals, faecal matter or domestic food waste detected and white represents contexts with no data. Contexts with micromorphological evidence of trampling are also noted and source of dung/animal indicator listed (C = cattle, LM = large mammal, MM = medium mammals).

micromorphology, since the only confirmed coprolitic remains were detected in context [251] from the inner section. The presence of faeces in context [251] is supported by the archaeobotanical evidence which contains the highest abundance of uncharred raspberry seeds, likely deposited within dung (e.g. Miller and Smart, 1984). The spatial distribution of faecal matter within Structure 2 could be related to (a) ruminant faecal matter being transferred into the inside of the roundhouse via trampling; (b) animal dung being used as hearth fuel and/or hide processing and (c) animals being kept within the structure.

The absence of evidence for ruminant faecal matter outside of the roundhouse based on M_{ex} would suggest that trampling could not be a source of ruminant faecal matter into Structure 2. Proxy comparisons further support this: for example, context [244] in M1 contains micromorphological evidence of trampling, yet no faecal signal is detected in the steroids. Steroid dung signals are also more prominent in the inner sections of Structure 2 (i.e. in M3 next to the central hearth structure), but if trampling was the key process then one would expect faecal matter to be widely distributed throughout the roundhouse.

Disentangling the causes of the stronger ruminant faecal signal near the hearth is difficult. It is possible this is related to dung storage, most likely for fuel, but we cannot rule out animal waste being produced in situ. If dung was kept close to the fire for ease of access, then this is likely to have accumulated on the floor surface surrounding the hearth. However, the main fuel identified from the macroplant analyses was charcoal and there is no clear evidence of burnt dung from the macroplant or micromorphological results or charred insect remains. Without geochemical analyses such as magnetics (e.g. Peters et al., 2004), XRF (e.g. Braadbaart et al., 2017) or phosphates (Macphail et al., 1997) on hearth deposits from Structure 2 it is difficult to eliminate dung as a fuel source. Based on the insect remains, the absence of charred dung and the

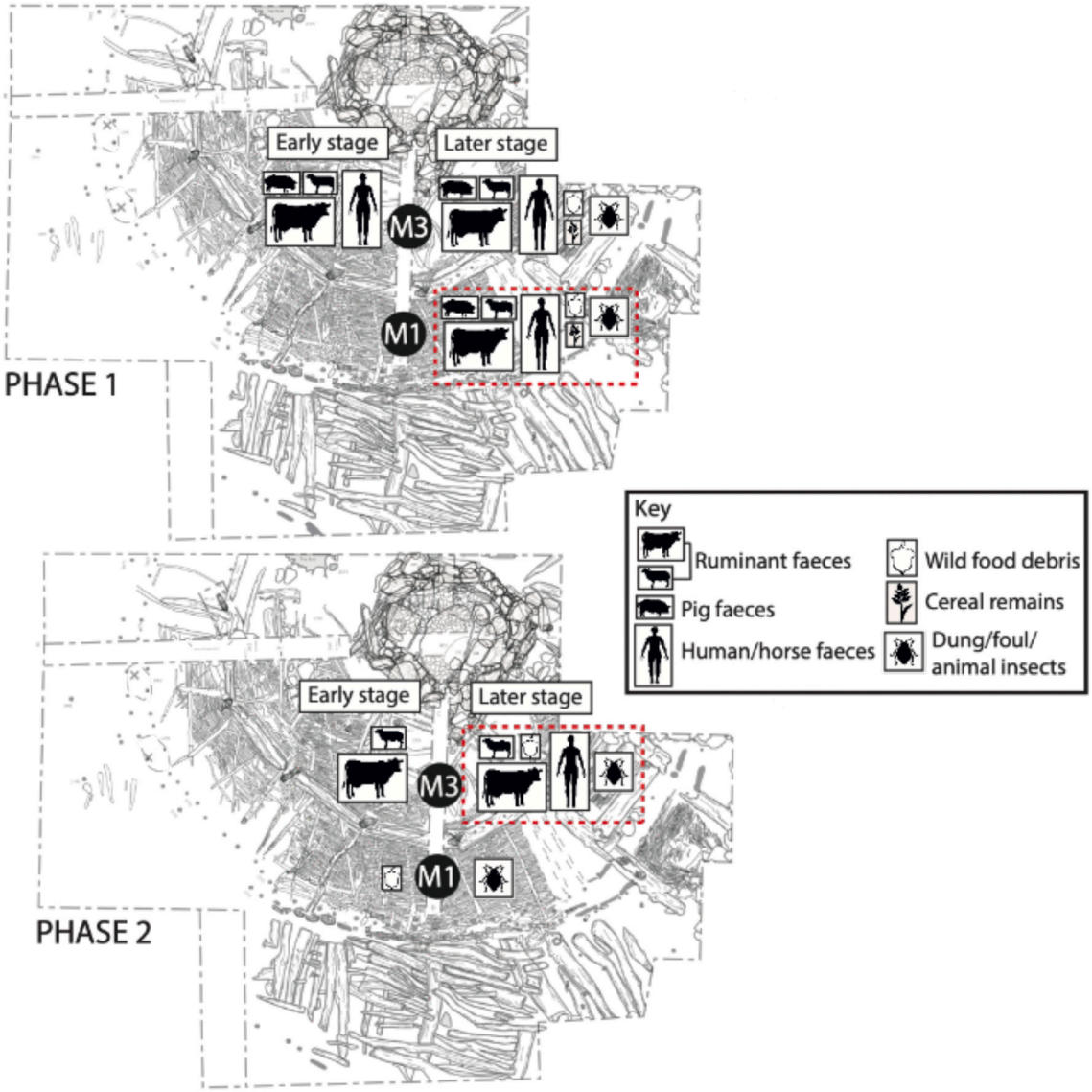

Fig. 8. Summary of multiproxy results highlighting differences in spatial and temporal use within Structure 2. Red boxes represent periods of multiproxy evidence for dominant dung/animal presence and household debris. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

wood charcoal in the macro-plant analysis, it is unlikely dung was a dominant source of fuel and therefore the faecal signal, within Structure 2.

The distribution of steroids most likely reflects the presence of designated livestock stalls within Structure 2. The more persistent faecal signal by the hearth could be explained by the deliberate placement of tethered animals proximal to the heat source to aid survival of the young or sick. Support for animal sheltering to improve the survival rates of new-born or unwell ruminants is evident from modern farming and veterinary studies as exposure is a key determinant of new-born mortality rates in wet and cold climatic conditions typical of Iron Age Scotland (e.g. Pollard, 2006; Hinch and Brien, 2014; Rawson et al., 1989). Placement by the hearth would also mimic the modern frost bite treatment for calves of rapid warming (Pelton et al., 2000).

4.3.2. Temporal patterns of use within structure 2

During Phase 1 of the occupation of the structure, the first source of faecal matter detected in both the inner and outer sections is horse and/or human and this indicates relatively foul living conditions, based on dung indicators across all proxies (Figs. 7 and 8), suggesting greater persistence or abundance of faecal matter. With the detection of charcoal and food debris, such as bones and seeds, this suggests the inner section of Phase 1 contains household debris.

In Phase 2 of the occupation, the bile acid results suggest that the faecal source changed to ruminants in both sections of the structure. The insect assemblage of dung-associated taxa, flies and lice, and the resemblance of the micromorphology of context [250] to stabling environments, as well as the digested berry seeds identified within the macrofossils, also point to animal activity and the accumulation of dung. The number of dung-associated taxa and concentrations of fly puparia present a strong argument for the presence of dung in a foul deposit, but it remains difficult to conclude the specific activities from the insect evidence alone as is the case with other studies with greater insect numbers (Forbes and Milek, 2014). The low concentrations and diversity of the dung insect community could be explained by regular removal and replacement of floor layers. The structure size would also limit the number of animals that could be housed (and thus the amount of dung produced) and the overall numbers of dung beetles would be reduced due to barriers to the outside created by walls (Smith et al., 2014). The presence of *Bovicola bovis* in the inner section of the

roundhouse highlights the complementary nature of faecal steroids and insect analyses, as the steroids confirm the presence of dung and insect indicator species refine the ruminant signal to confirm cattle and/or cattle hides were present.

4.3.3. Implications for use of structure 2 and daily Iron Age life

Possible functions of Structure 2 include space for sleeping, storage, food preparation, craft working and/or animal stalling (Pope, 2007). Our multiproxy analyses show there is no overwhelming evidence for sleeping in this particular structure, as insect concentrations are low with only one human louse identified and there is minimal structural evidence for bedding. Similarly, there is no conclusive evidence for craft working because there is no debris associated with this activity in the micromorphology and macroplant remains. Evidence for food preparation in the inner section of the roundhouse comes from small quantities of domestic debris within the macroplant remains and micromorphology, as well as the presence of cereal caryopses and chaff, which suggest small scale grain processing was likely to have been occurring. Storage within the outer section of structure 2 is possible, but there is no evidence for storage remains and the presence of the large hearth structure indicates storage is likely be a secondary rather than primary function of Structure 2.

The steroid evidence from Structure 2 suggests animals were present within the roundhouse, however, the combined evidence across all proxies does not support a long-term and/or intensive stabling environment. The absence of animal-derived steroids from M_{ex}, located outside of Structure 2, indicates that the roundhouse was used as a temporary or small scale area of human-animal cohabitation since we would expect to see a strong steroidal faecal signal outside the structure as a result of trampling and animal movement in and out of the roundhouse if this was a significant stabling environment. The ability to detect animal movement linked to stabling practises has been demonstrated using 5β-stanols analysed from the entrance of a stabling area in a modern experimental study of a reconstructed Iron Age roundhouse, which, unlike results from Structure 2, reported the entrance had similar sterol signatures to deposits located within the stable (Hjulström and Isaksson, 2009).

There is a lack of evidence for dedicated stabling structures or permanent 'byre-houses' (*sensu* Harding, 2004, 2009) in British Iron Age sites (Sørensen, 2007). The clearest evidence for co-habitation comes from outside Britain, from Nørre Tranders, Denmark (Nielsen, 2007). Interior stalling has been inferred from structural evidence and high phosphate levels at Woodend Farm in Dumfries and Galloway (Banks, 2000; Duncan, 2000) and excavations at Dun Vulan, South Uist suggest byre structures occurred within enclosures (Pearson and Sharples, 1999). The possible temporary presence of animals within occupied Iron Age structures has been identified from floor deposits after a rebuilding phase at Glastonbury Lake Village, England (Hill et al., 2018). The results from both Glastonbury Lake Village and Black Loch of Myrton (this study) indicate associations between animals and Iron Age roundhouses likely changed over time, reflecting variability of roundhouse usage. Whilst both structures may have been used as temporary small-scale co-habitations of humans and animals, there is no evidence to suggest they were permanent byre-houses.

The internal activity within Structure 2 based on our steroids, micromorphology and archaeobotanical remains (Fig. 8), follows Hingley's model of an active central area and peripheral outer area (Hingley, 1990) and supports the dominance of this model in Iron Age roundhouses in Northern Britain (*sensu* Hill, 1995; Pope, 2007). A peripheral, less frequently used outer area may also explain the abundance of flies detected in the outer section of the roundhouse since the reduced disturbance would facilitate fly larval pupation. Despite minimal evidence for activity in the outer section of the roundhouse compared with the inner section, micromorphological insights into floor cleaning and rebuilding in the outer section highlight the importance of maintaining the cleanliness of this area even under difficult waterlogged conditions. Hawkes (1994) suggested outer roundhouse areas were not characterised by inactivity but rather served important 'cleaner' functions, such as storing foodstuffs and firewood or sleeping. Interpretations of the multiproxy results across Structure 2 highlight efforts to frequently clean and maintain this roundhouse and support use as a shelter, likely with different primary functions depending on requirements over time.

5. Conclusions

Our study highlights the power of multiproxy approaches and the incorporation of steroids to advance insight into structure use, particularly when the sampling resolution facilitates characterisation of within-structure spatial and temporal patterns. Analyses of the Black Loch of Myrton's Structure 2 deposits provide evidence of floor cleaning and changes in use over time, demonstrating flexibility in roundhouse use over their short life cycle (*ca.* 30–40 years in this case). There is a more persistent faecal signal in the inner section compared with outer section of the roundhouse and this supports the 'active central area' roundhouse model (Hingley, 1990). In this case at Black Loch of Myrton, our data suggest small-scale temporary stabling within Structure 2, but likely only as a secondary function. Our results, however, highlight spatial complexity in roundhouse use as the outer area was less actively used and foul conditions persisted thus questioning the use of this space for 'clean conditions' (cf. Hingley, 1990).

Our application of steroids has successfully captured signals from short-lived/temporary pulses of faecal matter that are more difficult to extract from other traditional archaeological proxies. Our results also demonstrate steroids are particularly effective in archaeological settings with acidic soils, since they can identify the presence of animals where uncalcified bones do not preserve. Furthermore, faecal steroids provide valuable information about archaeological structures that have been subjected to the act of cleaning since they persist when visible indicators are removed or diminished.

The identification and characterisation of diffuse faecal input to the wetland settlement floor deposits within this study relies on analyses of both sterols and bile acids, supporting this combined analytical approach advocated by Bull et al. (2002) and Prost et al. (2017) to effectively overcome known issues relating to sterol ratio threshold values and help refine faecal source characterisations. The diffuse faecal steroid input associated with this Iron Age roundhouse has limited the resolution of faecal source characterisation compared with that achieved in more concentrated faecal remains (e.g. Prost et al., 2017; Harrault et al., 2019). However, the achieved source resolution is sufficient to advance understanding of human-animal interactions and cleanliness within the structure, and has benefited from further refinement through multiproxy comparisons.

The utility of incorporating steroid analysis is not restricted to wetland Iron Age structures, but is equally applicable to other periods and types of structures in different depositional settings. What is needed, however, are floors, inter-floor deposits, cleaning deposits or sealing layers contemporaneous with the abandonment of the structure. This study has highlighted the need for further experimental work focusing on diffuse faecal deposition in bulk occupation sediments to address questions raised about sensitivities of diagnostic ratios in such settings. A problem also highlighted here is where the controls should come from, especially within a settlement of several houses. An excavation of a small test pit outside the habitation area would seem most appropriate. As this study also shows, the combination of steroids with other proxies can help verify interpretations but may also raise new questions for investigation.

Author contributions

HM and ACGH designed the study in collaboration with AC, GC and AGB. HM conducted the primary lipid data analysis and interpretation with guidance from IDB and ACGH. KLD conducted the primary insect

data analysis and interpretation with NJW. JR and LR respectively conducted the primary macrofossil and micromorphology data. AC and GC facilitated sample collection and provided archaeological context for the study site. HM wrote the manuscript and all authors actively discussed the direction of the research and contributed to manuscript editing.

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