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Accessing the ephemeral using multiscale 3D microscopy of bone microwear

Naomi L. Martisius *

Department of Anthropology, The University of Tulsa, Tulsa, OK, USA
Department of Anthropology, University of California, Davis, Davis, CA, USA

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A B S T R A C T

Ancient people used perishable resources much as people do today. Such materials do not preserve well in the archaeological record, and so there are types of items that would have been made from animal skins or vegetal matter like clothing, footwear, and containers that are not present in many assemblages. Direct evidence for such items is often lacking, but indirect evidence in the form of microwear traces can be found on more durable materials, such as bone tools, that interacted with these less permanent items prior to deposition. In this study, 3D microscopic methods are applied to quantitatively evaluate traces from three perishable materials (semi-fresh animal skin, processed split leather, and dry bark) on experimental bone tools. Bones were scanned on two objectives (20x and 100x) with a confocal disc-scanning microscope and evaluated using 3D surface texture parameters. Quantitative measurements of the bone surfaces varied at the different scales, but material wear patterns were evident and indicative of the differences in properties of the three materials. This study indicates that the use of 3D microscopic methods on bone tools at multiple scales is useful for understanding how contact with perishable materials affects bone surfaces. This method has the potential to indirectly evaluate ephemeral resources that once came into contact with ancient bone artifacts and reconstruct some of the less tangible aspects of ancient human behavior.

1. Introduction

Humans exploit the resources available to them in any environment, and much of our material culture comes from organic sources such as plants and animals. The vast majority of these products are unlikely to survive over long stretches of time, though there is some direct evidence that human groups used wood and plant fibers as far back as the Middle and Late Pleistocene (Aranguren et al., 2018, Hardy et al., 2013, Hardy et al., 2020, Leroi-Gourhan, 1982, Nadel et al., 1994, Oakley et al., 1977, Schoch et al., 2015, Soffer et al., 2000, Thieme et al., 1985, Thieme, 1997). There is also indirect evidence for the processing of animal skins during similar time frames (Collard et al., 2016; d'Errico et al., 2003; Gilligan, 2007, 2010; Hallett et al., 2021; Krueger et al., 2019; Martisius et al., 2020a,b, 2022; Ocobock et al., 2021; Rots et al., 2015; Soressi et al., 2013; Wales, 2012). While direct archaeological evidence of artifacts made from perishable materials is minimal, these objects undoubtedly made up a substantial portion of ancient peoples' technological and social repertoire (Hurcombe, 2014). Animal skins or other animal parts could have been used for clothing, footwear, shelter, or containers. Some of these items likely had multiple components that may have come from a variety of other perishable materials. Plant fibers would have been ideal for making cordage or twine, as they are still used today. Unfortunately, it is more difficult to access these parts of the material past since objects made from plant and animal fibers rarely preserve in the archaeological record. The result is a gap in our understanding of ancient lifeways, social dynamics, and the day-to-day lived experience.

Lithic or bone tools, which preserve more readily, offer glimpses into missing perishable items and their function within ancient populations. The validity of these objects as proxies for processes involving perishable materials has been established (e.g., Marreiros et al., 2015, Semenov, 1964). Objects collected during ethnographic research can be compared with archaeological artifacts to develop ideas or inspiration for understanding material culture and processes in the past (Soffer, 2004, Stone, 2011b). While this line of research is insightful, there is a risk in equating the function of an object from one culture to a similarly shaped object from archaeological contexts (Stone, 2011b). Ancient peoples may have been making and using materials in ways that

* Address: Department of Anthropology, The University of Tulsa, Tulsa, OK, USA.
E-mail address: naomi-martisius@utulsa.edu.

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ethnographies cannot attest to. Common-sense analogies may also help to deduce the function of artifacts (Stone, 2011b). A particular tool may have certain properties that make it useful for a specific task or certain features of an artifact may point to a clear use. A perforated object such as a pendant would likely have been strung together or sewn onto some other material, possibly with twine made from plant material or a thin leather cord. Even so, certain objects, even those with distinct morphologies seemingly suited for specific tasks, may have been used for any number of purposes in manifold ways.

Microwear traces left from the use of an object can provide the necessary supplemental information for inferring an artifact's function. This has been accomplished using tribological principles for how two objects interact with each other (Bradfield, 2020a, Chomko, 1975, Lemoine, 1994). Based on material properties, specific traces produced during contact and friction of known materials should be reproducible and therefore identifiable on the surfaces of archaeological materials (Semenov, 1964). Experimental studies on a wide variety of materials have been undertaken to test these principles and build reference collections for assessing object functions (e.g., Buc, 2005, Campana, 1989, Lemoine, 1997, van Gijn, 2007). Varying experimental approaches attempting to replicate past human actions or focusing on assessing basic material principles are useful for evaluating potential artifact uses. Combined, these approaches have the potential to build a more complete understanding of materials present in the archaeological record (Marreiros et al., 2020).

Compared to stone, tools made from bone are ideal for assessing wear from perishable materials because they are they softer, accumulating well-developed microwear traces more readily than harder materials. They also tend to be used repetitively for long stretches of time to transform materials into something else entirely. Animal and plant material can be made into garments, sewn together, or woven into baskets, nets, and other useful objects. A great amount of work has been dedicated to assessing wear on bone from perishable materials at both low and high magnifications (Buc, 2005, 2011; Buc and Loponte, 2007; Campana, 1989; Christidou and Legrand, 2005; d'Errico et al., 1984; Griffitts and Bonsall, 2001; Legrand, 2007; LeMoine, 1991; Lemoine, 1997; Maigrot, 2003; Olsen, 1984; Shipman and Rose, 1988; Sidéra, 1993; Stone, 2011a, 2013; van Gijn, 2007; Legrand and Sidera, 2007). A key recognition based on this corpus of work is that volume deformation and alterations to the microtopography or microrelief of bone surfaces must be evaluated at different scales (e.g., Bradfield, 2022; Buc, 2011; Legrand and Sidera, 2007). A combined approach has helped microwear analysts to evaluate interacting materials and use their observations and understanding of material properties to assess how certain materials affect bone surfaces. Plant fibers, for example, have thick rigid cell walls that are often internally aligned. These can be reasonably predicted to cause wear on bone in a planar fashion with minimal edge rounding while producing structured and aligned striations (Stone, 2013, Stone, 2011a). On the other hand, animal fibers, rich in keratin and collagen, do not have rigid cell walls, are less orderly, and often incorporate particles from other sources. This combination may wear bone substantially by rounding the surface borders, invading all layers of the microtopography, while producing striations of varying shapes and sizes (Stone, 2013, Stone, 2011a). Even with a well-founded understanding of perishable material wear on bone, the inherent variation in all these materials can confound interpretations (Stone, 2013, Stone, 2011a).

While not a universal solution to this issue, a quantitative method that assesses measurable differences of certain surface features should limit any potentially objective decisions based on ambiguous features. 3D microscopy methods combined with the analysis of surface texture parameters has been used in recent years to more often measure the microtopography of stone tools (Evans and Donahue 2008, Evans and Macdonald 2011, Giusca et al. 2012, Ibáñez et al. 2018, Macdonald et al. 2018, Macdonald et al. 2019, Macdonald et al., 2020, Pedergnana et al., 2020, Stemp and Chung 2011, Stemp et al. 2015, Stevens et al. 2010, Werner 2018) but also bone surfaces (Bradfield, 2020b, d'Errico and Backwell, 2009, Lesnik, 2011, Martisius et al., 2018, Martisius et al., 2020a, Sinet-Mathiot et al., 2021, Vietti, 2016, Watson and Gleason, 2016). With the ability to quantify how differing perishable materials wear and alter bone microtopography, statistical modeling can fine-tune our understanding of the resulting differences and ultimately that of ancient artifacts used on such materials. To apply these methods to archaeological bone artifacts and have confidence in the results and interpretations, foundational methodological development is critical. For example, since bone is composed of organic materials subject to diagenesis and other post-depositional processes, it is not yet clear how these various processes affect the microtopography of bone and therefore the measurements used in surface texture analysis (Martisius et al., 2020a). Likewise, additional experimental analyses focused on assessing bone tools used with different actions on various contact materials and using a multiscalar approach is needed (Bradfield, 2022; Lesnik, 2011; Watson and Gleason, 2016; Legrand and Sidera, 2007).

This study builds on previous work that used a mechanical setup to control multiple variables and assess how bone wears due to contact with different perishable materials (Martisius et al., 2018). Surfaces were then analyzed using confocal disc-scanning microscopy and 3D surface texture analysis software. Here, some of the same analytical methods are used, but bones are worn in a less controlled way to see if similar results are obtained. If this is the case, bone surface analysis is one step closer to accurately assessing objects with unknown uses. Because different features are more apparent at different scales, applying this method using two different magnifications should provide varying results, not just in the values obtained across magnifications, but in the relationship of the materials to one another. Therefore, the multiscalar approach presented here will provide a more complete understanding of the raw material properties of bone, various perishable materials, and how they interact with each other.

2. Materials and methods

Five *Cervus elaphus* ribs from the same individual were obtained from a hunter. All ribs were cleaned and processed using a small amount of detergent and macerated for several months at the Zooarchaeology Laboratory at the University of California, Davis. After processing, the ribs were left to dry before any experiments were performed. The rough, irregular ventral end of the ribs were removed using a hammerstone. Ribs were further shaped through grinding against a dry ground stone until the modified, distal end of the experimental tool was spatula shaped. Grinding was most often performed at an oblique angle or transversely to the grain of the bone. Modifications were thorough, altering the working end of the bone so that overlapping grinding traces were visible over the entire surface area to be utilized. Two of the five ribs used in this study were reworked after their initial use to obtain additional samples. In total, nine tools were manufactured and used for this experiment.

Each experimental tool was used on one of three materials: semi-fresh skin (goat, *Capra hircus*), processed split leather (cow, *Bos taurus*), and dry bark (linden, *Tilia*). The goat skin was obtained from a farmer and came from a young animal. This skin was stored in a freezer for several months prior to its use in this experiment. The skin's membrane was removed and the internal surface was partially worked using other tools. No external abrasive substances were added to the skin at any point during the experiment. The bone tools made in the current study were used in the later stages of the process when the skin was beginning to dry out. The processed split leather was obtained from a department store and was soft and supple. The bark was obtained from Archeoshop, an experimental archaeology vendor, and came in strips from 1 to 2 cm wide.

One individual used three tools to work each material for 40 mins, most often with longitudinal and oblique motions. Tool use was not undertaken to achieve an altered state of the worked material, but rather to ensure that wear traces were well-developed on the surface of the distal tool portion. This was done because the tool shape used in this experiment may not have had a real-world application for each of the materials worked.

Once a tool was used on one of the three materials, the distal portion (~10 cm) was grooved and snapped off and curated for analyses. Unfortunately, one of the bones used on the skin was subsequently damaged and could not be further studied. Therefore, eight bone tools were analyzed (Table 1).

2.1. 3D surface texture analysis

Each bone tool was scanned at two magnifications with a confocal disc-scanning microscope (μsurf mobile, Nanofocus AG, Oberhausen, Germany), using a 20x (field of view = 0.8 mm^2) and 100x lens (field of view = 0.16 mm^2). The 20x lens was chosen to allow for a direct comparison of data with a previous study (Martisius et al., 2018). The 100x objective was selected to ensure sufficient contrast to the lower magnification used here. The numerical aperature values for the 20x and 100x objectives are 0.4 and 0.8, respectively. This difference should affect how the scans are acquired given that the different values relate to each objectives' ability to gather light and resolve fine detail, with higher values generally producing more accurate results (Calandra et al., 2019, Leach, 2011).

All artifacts were orientated with the object's longitudinal axis along the X-axis of the μsurf mobile and the distal end (tip) positioned at the extent of the X-axis. From this point, a grid was setup from a central (C) position in the active working area of each object, most often 3.375 and 1.35 mm proximally from the tip for the 20x and 100x, respectively (Fig. 1). Only one objective lens can be attached to the μsurf mobile at a time, so all scans on the 20x lens were taken prior to those on the 100x. Porous surface areas impeded systematic sampling, so five scans were captured within the main grid area at each objective were captured (Fig. 1). A small number of scans taken with the 100x lens were located within the scans taken at 20x (Fig. 2). Quality of scans were reviewed and accepted for further study if 95% or more of the surface points were measured (Martisius et al., 2018, Schulz et al., 2010, Schulz et al., 2013a). Lesser accuracy scans were re-measured by altering brightness, exposure, gain, or pitch values until 95% of the surface was captured.

Meshed axiomatic 3D models of each scan were made within MountainsMap Premium v. 7.4.8076 Analysis software by Digital Surf (Besançon, France) following standard procedures: leveling (LS-plane), outlier removal (edge and isolated outlier removal, with normal strength, and fill in points <225, noise removal), non-measured points fill in (smoothing method), and form removal using a polynomial of 2nd degree (Martisius et al., 2018, Martisius et al., 2020a, Schulz et al., 2010, Schulz et al., 2013a, Schulz et al., 2013b).

Nine surface texture parameters were selected for quantification using ISO 25178, isotropy, and scale-sensitive fractal analysis (SSFA).

Table 1
Experimental and confocal microscope scan details. Bone tools derive from one of five separate ribs, each used on one of three materials (dry bark, split feather, and semi-fresh skin), and had up to five usable scans taken at each objective (20x and 100x).

Bone tool	Material	Number of usable scans	
		(20x)	(100x)
1	Dry bark	5	5
2	Dry bark	5	5
3	Dry bark	5	5
4a	Split feather	5	5
4b	Split feather	4	5
4c	Semi-fresh skin	3	5
5a	Split feather	5	5
5b	Semi-fresh skin	5	5
5c	Semi-fresh skin	0	0

Parameters were chosen to be representative of nine main characterizing surface features (height, density, area, slope, peak sharpness, plateau size, volume, complexity, and direction) described in Schulz-Kornas et al. (2020) and based on previous experience assessing diagnostic alterations to bone microtopography as well as on the predicted differences in perishable materials (Fig. 3).

2.2. Statistical analysis of surface texture parameters

A subset of the nine surface texture parameters chosen for quantification were statistically analyzed using Bayesian modeling. Since many parameters are correlated, four relatively independent parameters were chosen based on pairwise scatter plots. Further, three of these parameters (*Sa*, *Sal*, and *Spc*) were used in a previous study employing the same statistical model allowing for a direct comparison of data (Martisius et al., 2018). An additional parameter (*Smr1*, upper material ratio) used in the Martisius et al. (2018) study could not be included here because it was unobtainable at both 20x and 100x magnifications. Instead, A fourth parameter (*Smr*) representing the same characterizing feature is used here.

Bayesian modeling was employed following previous protocols (Martisius et al., 2018, Martisius et al., 2020a, Sinet-Mathiot et al., 2021). Three increasingly complex statistical multivariate mixed models of the form $Y = XB + ZU + E$ (see Amemiya, 1994) were employed for the four log-transformed parameters from 77 meshed axiomatic 3D models making **Y** a 4x77 matrix (Table 2). M0, the empty model with no fixed effects **XB**, includes random effects **ZU** and residual error **E**. The idiosyncratic effects of individual *ribs* and *measurement locations* are represented by **ZU**, where **U**, an 82x4 matrix, contains a column of 5 unique *rib* effects and 77 *measurement location* effects and a second column of the four surface texture parameters. **Z** is a 77x82 matrix of zeros and ones indicating the *rib* and *measurement location* of each scan. **E** is a 77x4 residual matrix. The dimensions of **XB** are dependent on the number of fixed effects in the model. M1 includes the fixed effects *material* and *magnification*, while M2 also includes the interaction of *material* and *magnification*. Leave-one-out cross-validation (LOO) scores of the models (Vehtari et al., 2017) found the most complex design (M2) to generate model predictions best (Table 2), so **B** in M2 contains 6 fixed effects for each of the four surface texture parameters resulting in a 6x4 matrix. **X** is a 77x6 matrix of zeroes and ones. Therefore, M2 is a multilevel, multivariate Bayesian model that includes a pairwise interaction of fixed effects (*material* and *magnification*), random effects (*rib* and *measurement location*), and *error*.

A goodness of fit check was applied to ensure that *rib* and *measurement location* random effects were adequately modeled using multivariate Gaussian distributions. For additional model details see Martisius et al. (2018). Effects were estimated using a Hamiltonian Markov-chain Monte Carlo method, using the library rstan version 2.21.2 (Stan Development Team, 2021) of the statistical computing language R version 4.1.0 (R Core Team, 2021) and allowed a 2000-iteration warm-up for four chains generating 1000 parameter samples per chain resulting in 4000 posterior samples for inference. Scaled and squared Mahalanobis distances were examined between observations and predicted values to check for goodness of fit and distances were compared to theoretical quantiles of the F-distribution (Roth, 2013).

3. Results

A comparison of nine surface texture parameters with different fields of view reveals different values for most parameters and material types (Fig. 4). This was expected, given the vastly different scales at which the measurements were taken (Fig. 5). In general, *Sa*, *Sal*, *Sda*, and *Vvv* indicate smaller values at higher magnification, while *Sdq*, *Spc*, and *FD* are larger, trends consistent with each parameters' unit of measurement. *Smr* and *IsT* exhibit overlapping variation at both scales, also consistent with their measurement units, which are expressed as percentages.

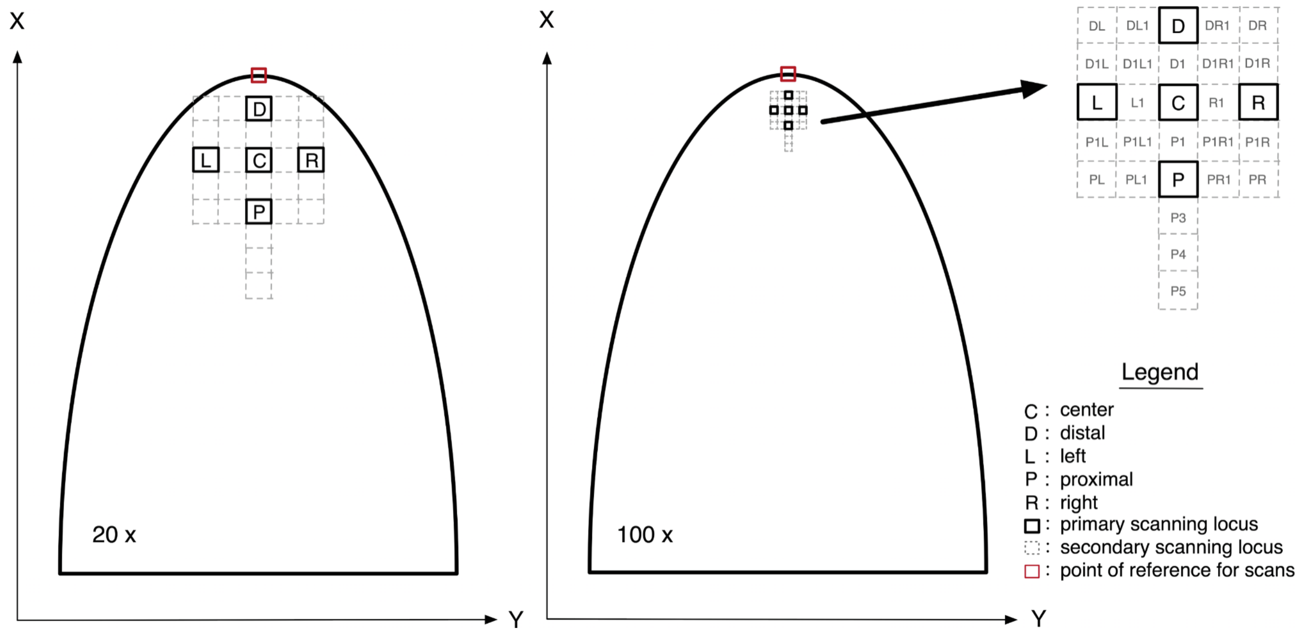

Fig. 1. Exampfles of sampflfing method used for obtafinfing surface texture scans on the experfimentafl bone toofls at flocatfions C (center), D (dfistafl), P (proxfimafl), L (fleft), and R (rfight). Addfitfionafl scanned flocatfions are represented by dashed boxes. Scanned areas are 0.8 and 0.16 mm^2 at 20x and 100x magnfifficatfion, respectfivefly, but may not be depficted to scafle here.

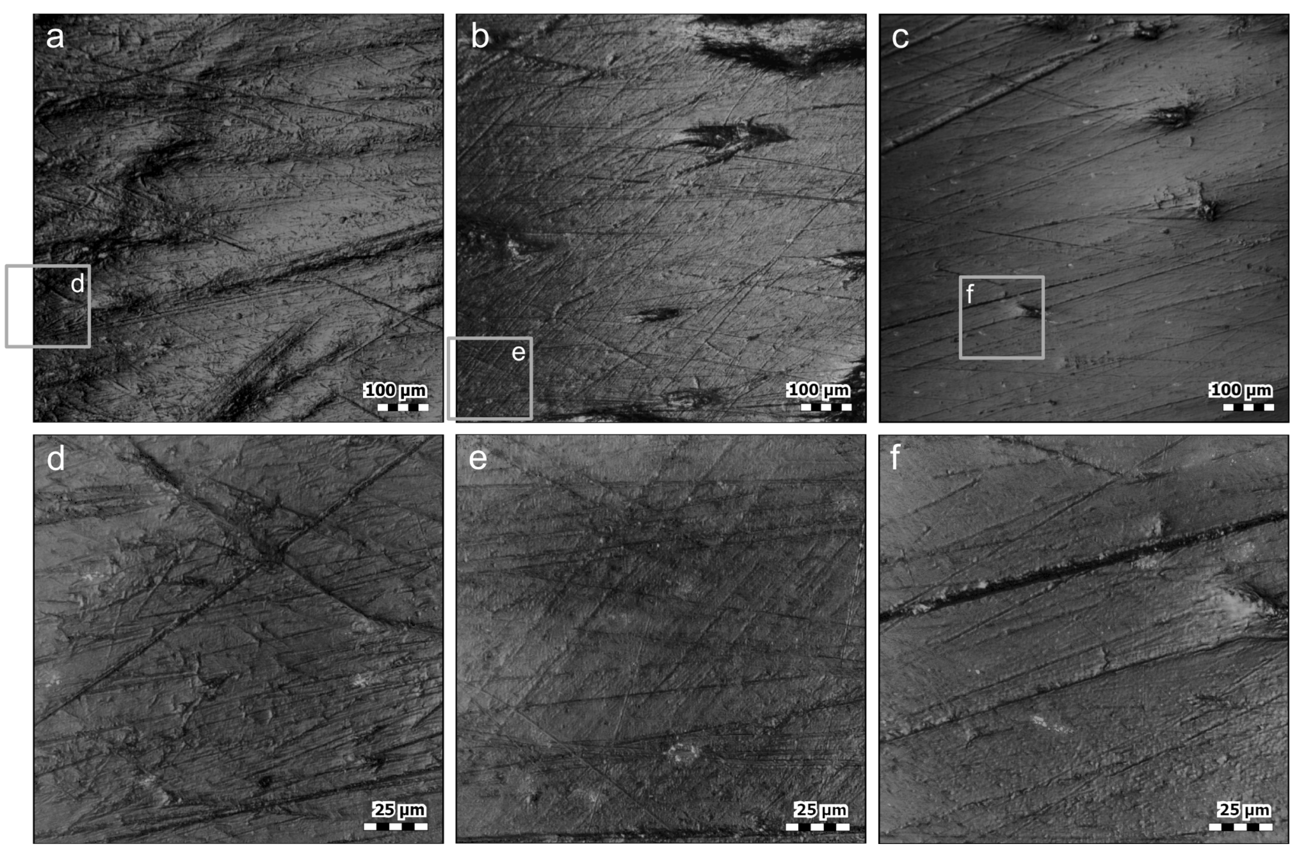

Fig. 2. 2D fintensfity fimages of bone surface mficrotopography at 20x (a-c) and 100x (d-f) magnfifficatfion. Bone toofls 3 (a and d), 5b (b and e), and 4b (c and f) were used on dry bark, spflfit fleather, and semfi-fresh skfin, respectfivefly.

Further, vaflues on the 20x flens exhfibfit wfider ranges for most of the parameters and more overflap fin vaflues than at 100x.

When comparfing the reflatfionshfip fin the vaflues of the materfiafl types, some parameters (*Spc*, *Sda*, *Smr*, and *FD*) findficate a sfimfiflar trend across the two scafles. At both magnfifficatfions, the bones worn on bark exhfibfit the hfighest vaflue fin *Spc* meanfing that the peaks of the surfaces are sharper, whereas both skfin- and fleather-worn bones have more rounded peaks (Ffig. 4). Further, the fleather-worn surfaces are the most rounded,

Parameter	Description	Functional group	Standard	Unit	Low	High
Sa	Arithmetic mean height or mean surface roughness	Height	ISO 25178	μm		
Sal	Autocorrelation length (s = 0.2)	Density	ISO 25178	μm		
Sda	Closed dale area	Area	ISO 25178	μm^2		
Sdq	Root-mean-square gradient	Slope	ISO 25178	no unit		
Spc	Arithmetic mean peak curvature	Peak sharpness	ISO 25178	$\frac{1}{\mu m}$		
Smr	Areal material ratio, bearing area at given height (c = 1 μm under the highest peak)	Plateau size	ISO 25178	%		
Vvv	Void volume of the valleys at a given material ratio (p = 80%)	Volume	ISO 25178	$\frac{\mu m^3}{\mu m^2}$		
FD	Fractal dimension	Complexity	SSFA	$\frac{1}{\mu m^2}$		
IsT	Texture isotropy	Direction	Isotropy	%		

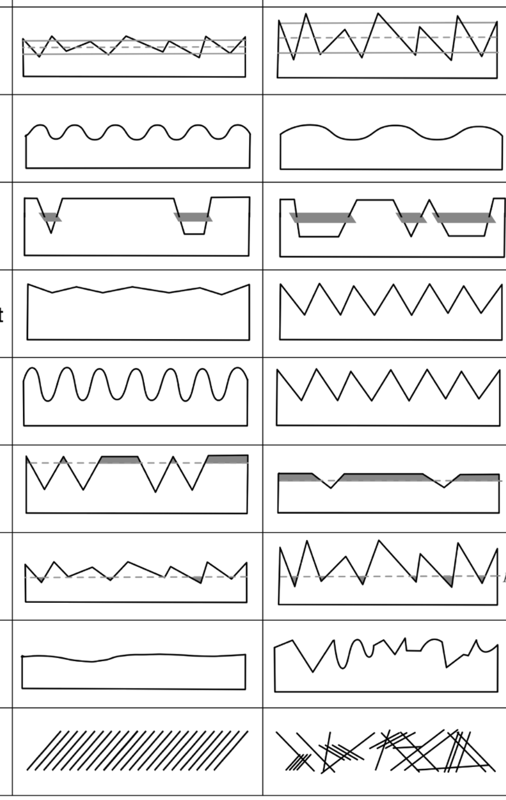

Fig. 3. Surface texture parameter descrfiptfions wfith functfionafl group, standard, and unfits of parameters accordfing to ISO 25178, texture dfirectfion, and scafle-sensfitfive fractafl anaflysfis (SSFA). Orthographfic depfictfions findficate exampfles of surfaces wfith flow and hfigh vaflues (adapted/modfiffied from Kafiser et afl. (2016) and Martfisfius et afl. (2018, 2020a, Sfinet-Mathfiot et afl., 2021)).

Table 2
Modefl effects fincfludfing thefir representatfion fin each modefl's desfign space and LOO expected flog predfictfive densfity (eflpd) and standard error (se) dfifferences reflatfive to M2.

Model	Effects	Design matrix	Δ elpd	Δ se
M2	Ffixed + random + pafirwfise finteractfion	Materfiafl + magnfifficatfion + materfiafl*magnfifficatfion + rfib + measurement flocatfion + error	0.0	0.0
M1	Ffixed + random	Materfiafl + magnfifficatfion + rfib + measurement flocatfion + error	26.8	12.1
M0	Random	Rfib + measurement flocatfion + error	55.3	18.8

but aflso exhfibfit the most varfiatfion. Materfiafl type worn exhfibfits the same reflatfionshfip across scafles for *Sda*. At both magnfifficatfions skfin-worn surfaces have the flowest vaflues findficatfing a smaflfler cross-sectfionafl dafle area fin the flower portfion of the surfaces, whfifle those worn by fleather have the flargest dafle area (Ffig. 4). Interestfingfly, both *Smr* and *FD* findficate that skfin-worn surfaces have some of the hfighest vaflues across the scafles meanfing that such surfaces exhfibfit the flowest surface reflfiefs whfifle havfing the most compflex surfaces, though the flarge amount of varfiatfion fin the fleather sampfle at 100x findficates that some of the fleather-worn surfaces have the flowest surface reflfiefs (>*Smr*). Further, bones used wfith bark exhfibfit the hfighest surface reflfiefs (<*Smr*), whfifle those wfith fleather have some of the fleast compflex surfaces (<*FD*) across both magnfifficatfions.

The ffive other parameters (*Sa*, *Sal*, *Sdq*, *Vvv*, and *IsT*) show sflfight dfifferences at the two magnfifficatfions. For *Sa*, skfin-worn surfaces have the flowest vaflue, or fless surface roughness, on the 20x, whfifle bone used on fleather fis flowest on 100x (Ffig. 4). The bark-worn surfaces appear to be some of the roughest at both magnfifficatfions, aflbefit the fleather-worn surfaces aflso exhfibfit hfigher vaflues at 20x. The pattern observed for *Vvv* fis very sfimfiflar to that of *Sa*, whfich findficates that the surfaces that are most rough (>*Sa*), such as the bark-worn surfaces, aflso have the flargest voflume fin the vaflfleys of the surface (>*Vvv*). For *Sal*, bones used on fleather have the flowest frequency surfaces (hfigher vaflues) on the 20x, but on the 100x, both fleather- and bark-worn surfaces exhfibfit sfimfiflarfly flow frequency surfaces (Ffig. 4). On the other hand, bones used on skfin and bark exhfibfit hfigher frequency surfaces at 20x, whfifle those used on skfin are nearfly fisoflated wfith the hfighest frequency surfaces at 100x. In generafl, the pattern observed for *Sdq* fis very sfimfiflar to that of *Spc* (descrfibed fin the prevfious paragraph) at both magnfifficatfions, whfich findficates that the surfaces wfith the sharpest peaks (>*Spc*), such as the bark-worn surfaces, aflso have the steepest surface sflopes (>*Sdq*). However, skfin-worn surfaces show a sflfightfly dfifferent pattern at the two scafles. At flower magnfifficatfion, bones used on skfin and fleather are sfimfiflar fin havfing graduaflfly sflopfing surfaces, whfifle at hfigher magnfifficatfion, skfin-worn surfaces are more flfike those used on bark and have steeper sflopes. For *IsT*, bones used on bark have the flowest vaflues at 20x but some of the hfighest vaflues at 100x, whfich findficates a more anfisotropfic surface at flower magnfifficatfion and fisotropfic at hfigher magnfifficatfion (Ffig. 4). An opposfite trend fis observed for surfaces used on skfin. Overaflfl, the varfiatfion exhfibfited fin the *IsT* vaflues fis a mfixture of structured and fless structured surfaces for each materfiafl type as findficated by the flarge amount of varfiatfion at both scafles for most materfiafl types, though most vaflues are fless than 50% findficatfing anfisotropfic surfaces

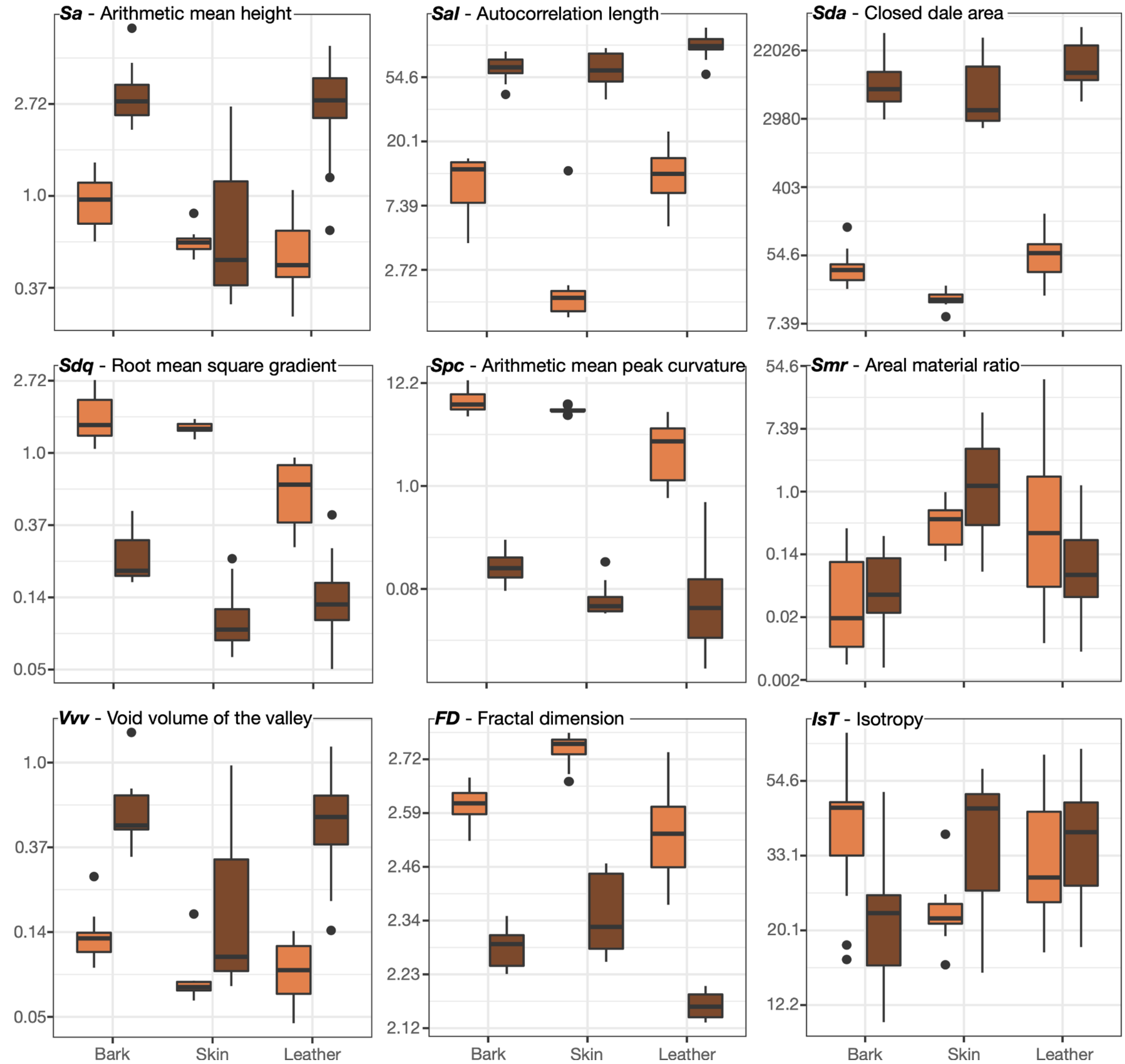

Fig. 4. Boxpflots of surface texture parameter vaflues for each materfiafl type at 20x magnfifficatfion (dark brown) and 100x magnfifficatfion (orange). Y-axes are on the flog scafle, but tfick flabefls are fin orfigfinafl measurement unfits to fimprove finterpretabfiflfity between scafles. (For finterpretatfion of the references to coflor fin thfis ffigure flegend, the reader fis referred to the web versfion of thfis artficfle.)

overaflfl.

Whfifle an evafluatfion of scatterpflot matrfices aflso reveafls the same trends as the boxpflots for the findfivfiduafl surface texture parameters at dfifferent scafles (Ffig. 4), scatterpflots are usefufl for evafluatfing more than one parameter at a tfime (Ffig. 6). In addfitfion, these pflots fincorporate modefl effects exhfibfited by the flarge 95% posterfior contours for the estfimated means of each pafired parameter (Ffig. 6), whfich gfiven the fidfiosyncratfic varfiatfion fin bone surfaces, fis fimportant when makfing finferences from thfis type of data (mficroscopfic measurements). Further, fincorporatfing the mufltfivarfiate modefl predfictfions reveafls sets of parameters that appear to be especfiaflfly usefufl for dfistfingufishfing materfiafl type wear on bone artffacts.

On the 20x flens, the 95% posterfior contours for bones used on skfin findficate mfinfimafl overflap fin vaflues for *Sa* (fless rough) and *Smr* (flowest surface reflfiefs) on thfis scafle compared to the other materfiafls (Ffig. 6). Whfifle there fis overflap fin most other modefl predfictfions, fleather-worn surfaces have some of the flower frequency surfaces (hfighest *Sal* vaflues) at flower magnfifficatfion and those used on bark exhfibfit some of the hfighest vaflues for *Spc* (fless smooth). These modefled comparfisons findficate that *Sa*, *Spc*, and *Smr* are the more usefufl parameters for dfistfingufishfing materfiafl type at flower magnfifficatfion.

At 100x, there fis better separatfion fin the 95% posterfior contours for the estfimated means of some of the parameters (Ffig. 6). At thfis magnfifficatfion, the modefl predfictfions for *Sal* findficate that bones used on skfin are compfletefly dfistfingufishabfle from the other materfiafls and have the hfighest frequency surfaces (<*Sal*). In addfitfion, the modefl predficts that fleather-worn surfaces have mfinfimafl overflap fin vaflues usfing *Spc* compared wfith other materfiafls, whfich findficates that bones used on fleather exhfibfit the most rounded peaks (<*Spc*). In fact, the modefl predfictfions for the comparfison *Sal*:*Spc* findficates that aflfl three materfiafl types are cflearfly dfistfingufished at 100x (Ffig. 6b).

4. Discussion

Archaeoflogficafl bone toofls exhfibfit a serfies of traces findficatfive of thefir flong flfife-hfistorfies. These are acqufired fin a compflex paflfimpsest as the toofl fis made and then used. Manufacturfing traces tend to be observabfle at a macroscopfic scafle, wfith detafifls more cflearfly vfisfibfle at the mficroscopfic flevefl (e.g., Campana, 1980, Lemofine, 1994, Newcomer, 1974). Sfimfiflarfly, traces from an object's use can be seen at mufltfipfle scafles, but hfigher magnfifficatfions (generaflfly > 50x usfing optficafl mficroscopy) are needed to observe any unfique features that may be findficatfive of the materfiafl on whfich the bone was used (e.g., Bradffiefld, 2022; d'Errfico, 1993; Lemofine, 1994; Marrefiros et afl., 2015; Semenov, 1964). The

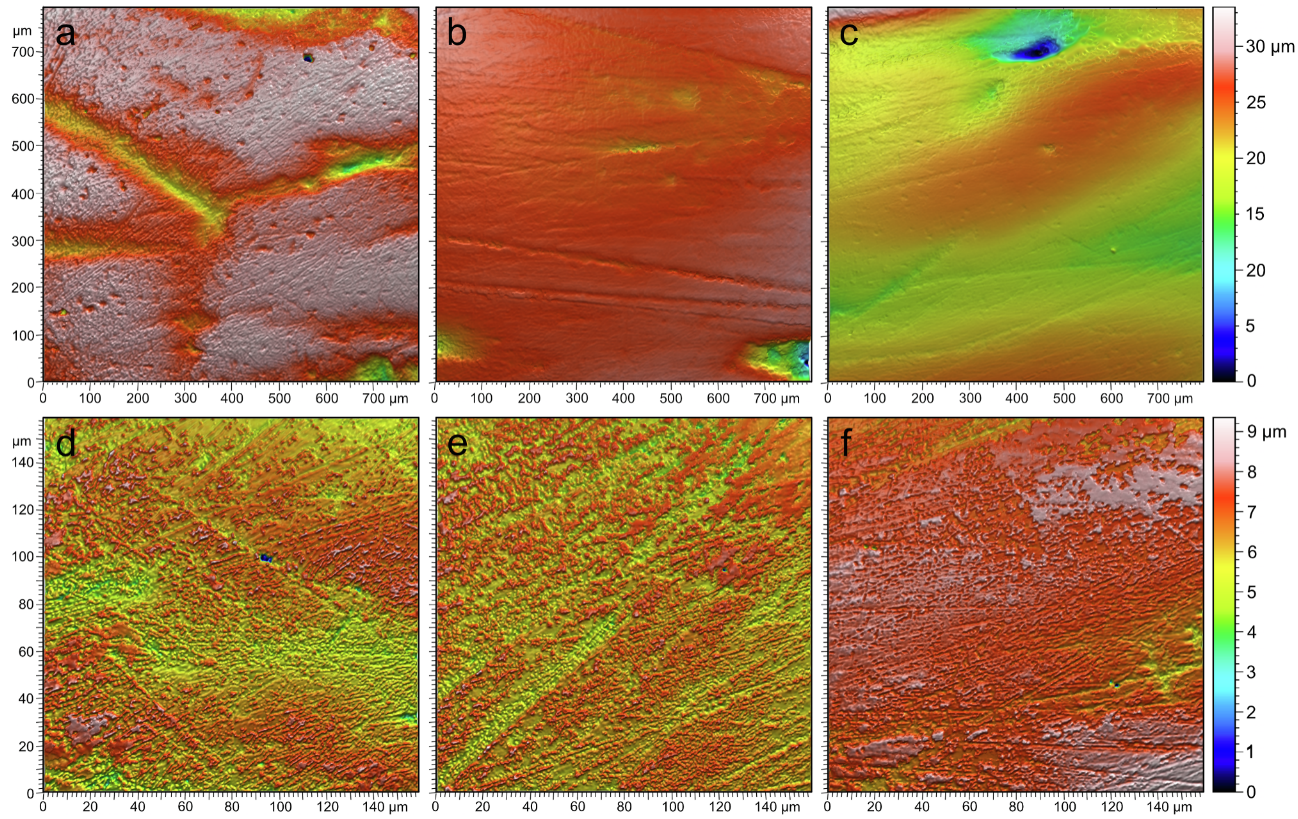

Fig. 5. Meshed axfiomatfic 3D modefls of surfaces representatfive of the three types of perfishabfle materfiafl wear tested fin thfis study (a and c, dry bark; b and e, semfi-fresh skfin; c and f, spflfit grafin fleather) at 20x magnfifficatfion (a-c; 0.8 mm^2) and 100x magnfifficatfion (d-e; 0.16 mm^2). Coflor of surfaces correspond to hefight on the z-axfis fin upper rfight-hand corner.

degree of magnfifficatfion depends on mficroscope settfings and contact materfiafls, but objectfives wfith a ffiefld of vfiew approxfimatefly or narrower than 2 mm2 are generaflfly needed to assess mficrowear traces (e.g., Bradffiefld, 2015, 2022; Buc, 2011; Marrefiros et afl., 2015). Once a bone toofl fis dfiscarded, fit may accumuflate addfitfionafl surface modfiffiecatfions characterfistfic of dfifferfing taphonomfic processes (e.g., Behrensmeyer, 1978, Fernandez-Jaflvo and Andrews, 2016, Ffisher, 1995). Dfisentangflfing traces of dfifferfing magnfitudes and dfimensfions produced durfing varfious processes fis essentfiafl for any meanfingfufl finterpretatfion of ancfient human behavfior. Thfis study fis focused on evafluatfing experfimentaflfly produced mficrowear traces, and the resuflts findficate that there are quantfiffiabfle 'sfignatures' for dfifferent materfiafls that change at the scafle of measurement.

The dfifferent resuflts obtafined at the dfifferent ffiefld s of vfiew have fimpflficatfions for comparatfive mficrowear studfies. Dependfing on the quantfitatfive parameter used, cross-scafle comparfisons are not advfisabfle, even fif generafl trends are compared rather than specfiffic vaflues. It has recentfly been shown that the objectfive and numerficafl aperture (na) used for scannfing surfaces finffluences ISO 25178 parameters, even when used at the same magnfifficatfion (Caflandra et afl., 2019). Dfifferences fin the na vaflues of the 20x and 100x objectfives used fin the current study may contrfibute to the dfifferences fin measurements. It fis flfikefly that the flow na vaflue (0.4) of the 20x flens contrfibuted to the greater dfistrfibutfion of vaflues at that scafle. Whfifle fit fis flfikefly that na finffluences the vaflues obtafined, fit fis unflfikefly to flead to compfletefly dfifferent trends for the surfaces worn by dfifferfing materfiafls. Caflandra et afl. (2019) found that an objectfive wfith a hfigher na vaflue consfistentfly produced sflfightfly flower *Sa*, *Sdq*, and *Vvv* vaflues and sflfightfly hfigher *Sal* vaflues. The dfifferences fin objectfives for *Smr* and *Str* (comparabfle to fisotropy parameter *IsT*) were varfiabfle, but thfis resuflt fis not finconsfistent wfith the unfit of measurement for these parameters (scafle of 0 to 1). In fact, most of the parameters tested wfith the two objectfives shfifted fin one consfistent dfirectfion (Caflandra et afl., 2019). The dfifferent vaflues at the dfifferent magnfifficatfions obtafined fin the present study for *Sa*, *Sal*, *Sdq*, *Vvv*, and *IsT* do not foflflow the same trend. Instead, the flargest dfifferences are most often reflated to one of the three materfiafl types, skfin. Further studfies usfing the same na on objectfives wfith dfifferent magnfifficatfions fis needed to descrfibe the dfifferences more cflearfly at the two scafles, but crucfiaflfly, fit fis flfikefly that the trends observed at the dfifferent magnfifficatfions are reafl and findficatfive of materfiafl wear and not due to mficroscope settfings.

Mufltfiscafle studfies of measured surface topographfies of bone toofls are stfiflfl fin earfly stages of research but have been successfufl fin reflated ffiefld s fincfludfing pafleontoflogy and for the anaflysfis of stone toofls (e.g., Brown et afl., 2018, Macdonafld et afl., 2020, Stemp, 2014, Stemp et afl., 2010). Two prevfious studfies usfing flaser scannfing confocafl mficroscopy on bone toofls showed that scafle sensfitfive fractafl anaflysfis, often usfing area-scafle fractafl compflexfity (*Asfc*), was usefufl for dfistfingufishfing dfifferent types of wear on bone (Lesnfik, 2011, Watson and Gfleason, 2016). Nonethefless, sfimfiflar anaflyses have not been appflfied more frequentfly. On the other hand, quaflfitatfive mficrowear anaflysfis on bone fis commonfly appflfied at mufltfipfle scafles (e.g., Bradffiefld, 2022; Buc, 2011; Chrfistfidou and Legrand, 2005; Grfiffiftts, 2006; Legrand, 2007; Lemofine, 1994; Stone, 2013; van Gfijn, 2007; Legrand and Sfidera, 2007). To better dfistfingufish dfifferent types of materfiafl wear, tradfitfionafl bone mficrowear anaflysts often dfistfingufish between and descrfibe dfifferfing features of the mficrotopography and the mficroreflfiefs (e.g., Buc, 2011, Legrand, 2007), whfich may present very dfifferentfly. For exampfle, Buc (2011) descrfibes the mficrotopography of experfimentafl awfls used for pfiercfing fresh skfin as efither homogeneous or heterogeneous, dependfing on the bone object, and wfith crossfing strfiatfions. Mficroreflfiefs are descrfibed as reguflar wfith rough eflevatfions that exhfibfit deep, smooth strfiatfions (Buc, 2011). The descrfiptfions of the observed features cflearfly change wfith the scafle of

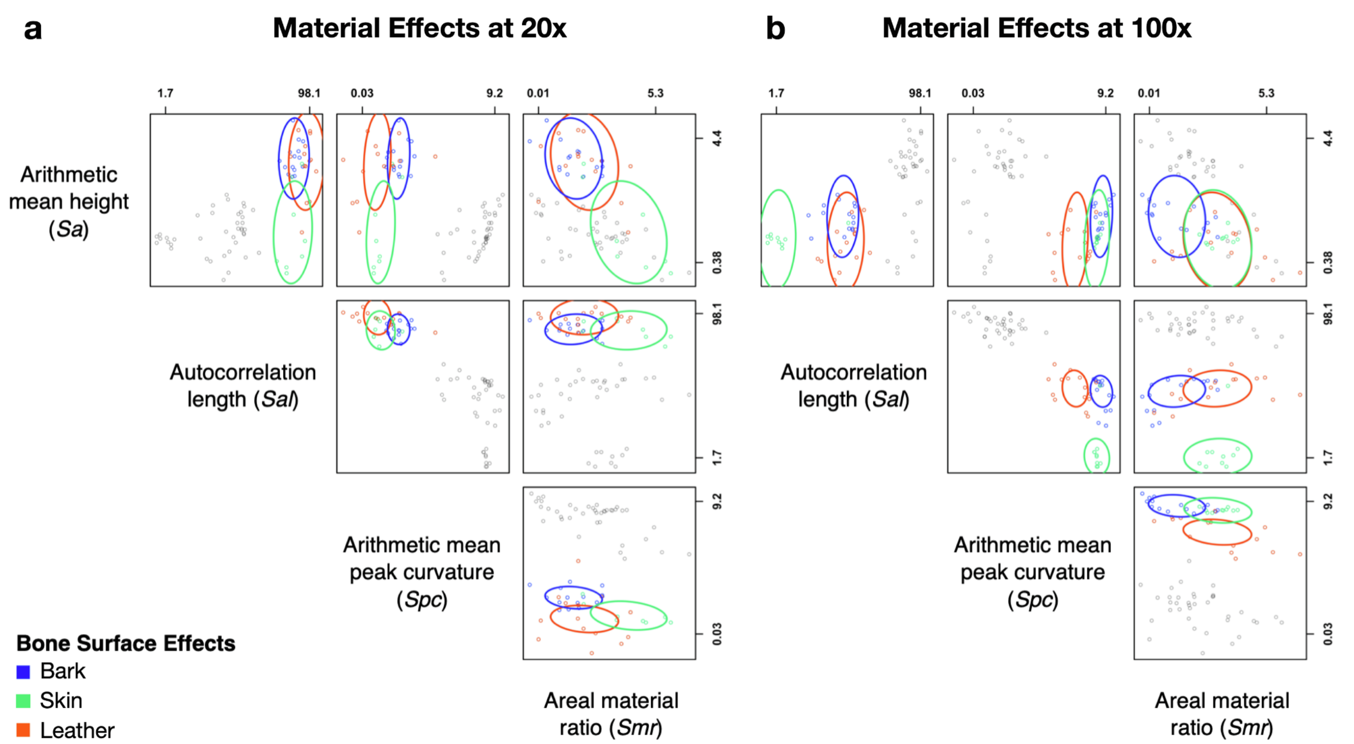

Fig. 6. Scatterplot matrices at 20x and 100x magnification. Plots show all observations displayed in the pairwise space of parameters: surface roughness (*Sa*), autocorrelation length (*Sal*), peak curvature (*Spc*), and areal material ratio (*Smr*). Colored observations are representative of material type (blue: bark; green: skin; orange: leather) at (a) 20x and (b) 100x magnification and ellipses show the model predictions of the mean for each pair of parameters. Axes are on the log scale, but tick labels are in original measurement units and placed at the 5th and 95th percentiles. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

observation. While comparison between traditional qualitative features and quantitative surface parameters do not describe the exact same features, the differences at the different scales in the current study are likely related to the observations made by Buc (2011) and many others (e.g., Bradfield, 2015, 2022; Christidou and Legrand, 2005; Griffitts, 2006; Legrand, 2007; Lemoine, 1994; Stone, 2013; van Gijn, 2007; Legrand and Sidera, 2007).

In this study, skin exhibits the greatest differences at the two scales as indicated by the different trends in several parameters (*Sa*, *Sal*, *Sdq*, *Vvv*, and *IsT*). At lower magnification, skin appears to produce the smoothest surfaces (<*Sa*) with the least amount of volume in the valleys of the surface microtopography (<*Vvv*), while at higher magnification, leather-worn surfaces exhibit these qualities, albeit bones used on skin are within the total range of the values in the leather sample. A similar pattern is observed for *Sdq*, which indicates that skin-worn surfaces are the least sloped at lower magnifications, but along with bones used on bark, have steeper slopes at higher magnification. The overall pattern, which includes parameters that indicate a similar trend across magnifications (<*Sda*, small cross-section of dales, >*Smr*, low surface reliefs, >*FD*, greater complexity), is consistent with skin being more abrasive than the other materials, therefore resulting in increased attrition and thorough modification of the microtopography across a large area (LeMoine, 1991). At the same time, the abrasive quality of skin, possibly aided by exterior inclusions, likely resulted in slightly deeper and steeper furrows adding to its roughness at higher magnification.

The values of *Sal* produced by the different perishable materials and their relationship to each other is also different across magnifications. *Sal* measures the wavelength of the surface, so those that are nearly flat should produce a low value (shorter wavelength). In general, lower magnification scans exhibit higher values because larger features such as pores or the remnants of grinding traces are a component of many of these scans. At this lower magnification, the leather-worn surfaces preserve many of these larger features. In general, scans at higher magnification produce *Sal* values that are lower because these scans do not fully capture the large features. Even so, the bones worn by leather and bark, to an extent, retain features from the manufacturing process that were not fully worn down, while skin-worn surfaces are nearly flat. This result is consistent with skin being the most abrasive of the tested materials, resulting in thorough modification of not only the micro-topography but of the microreliefs as well.

The semi-fresh skin and the processed split leather used in this study are two different states of the same material type, so there should be similarities in their effect on bone. Stone (2013, 2011a) produced a tribological model for perishable material wear on bone after compiling data from various sources (e.g., Buc, 2005, Buc and Loponte, 2007, Christidou and Legrand, 2005, Griffitts, 2006, Legrand and Sidera, 2007, Lemoine, 1997, Maigrot, 2003), and confirmed that several features were indicative of animal material wear: invasive polish, rounded microtopography, and variably present striations that are irregular in organization. Interestingly, many of the surface texture parameter values for the two different states of animal skin in the current study indicate different values. However, more similarities are evident at lower magnification, which is the scale at which the features described by Stone (2013, 2011a) are all clearly visible. Of note are the similarities in *Sdq*, *Spc*, and *IsT*, which indicate that the two animal skin materials produce gradually sloping and rounded surface peaks and surfaces that are variably directionally oriented (ie., variable striations). In this case, these three surface texture parameters clearly measure the same features that qualitative microwear studies on bone have often described.

While skin and leather exhibit a few similarities at lower magnification, their unique material properties result in some very clear differences (Buc, 2011, LeMoine, 1991). As previously described, surface roughness (*Sa*) exhibits differing trends at the two scales with skin producing the smoothest surfaces at lower magnification, while leather does so at higher magnification. This indicates that skin affects the entire microtopography of the bone surface, whereas fully processed leather appears to have thoroughfly smoothed some parts of the surface (most often the upper reflfiefs). Thfis dfifference together wfith the consfistent resuflt of fleather havfing some of the hfighest *Sal* and *Sda* vaflues (flonger waveflength, flarger dafle area) and the flowest *Spc*, *Sdq*, and *FD* vaflues (rounded peaks, graduafl sflopfing, fless compflex) on both scafles findficates that fleather exhfibfits a poflfishfing effect where the materfiafl penetrates the mficrotopography of the surface roundfing the peaks but does not resuflt fin thorough attrfitfion of the bone surface.

Bones used on bark exhfibfit a serfies of characterfistfic features that dfiffer from the anfimafl materfiafls. At flower magnfifficatfion, bark produces surfaces that are more anfisotropfic (<*IsT*, more dfirectfionaflfly orfiented), whfifle skfin-worn surfaces are the most anfisotropfic at hfigher magnfifficatfion. Interestfingfly, bark appears to exhfibfit some of the most fisotropfic surfaces at hfigher magnfifficatfion. Due to bark's thfick, rfigfidfly aflfigned ceflfl waflfls (Stone, 2011a), the presence of organfized strfiatfions or an anfisotropfic surface fis expected as fis observed at flower magnfifficatfion. When zoomed out, the flarger pattern fis flfikefly findficatfive of the overaflfl organfizatfion of the bark ceflfluflar structure, but when zoomed fin, a more randomfized pattern mfight be caused by the firreguflar movement of sfingfle ceflfl structures or a smaflfl coflflectfion of them. The resuflt that skfin produces anfisotropfic surfaces at hfigher magnfifficatfion fis surprfisfing gfiven the softer ceflfl structures that are fless organfized (Stone, 2011a). Though the skfin-worn surface vaflues are flow, they stfiflfl faflfl wfithfin the range of those used on fleather at both scafles and skfin at 20x. The flower vaflues may be the resuflt of a smaflfler sampfle sfize avafiflabfle for bones used on skfin.

The other characterfistfic resuflts for bark, hfigher *Sa, Sdq, Spc,* and *Vvv* (rougher, steeper, sharper peaks, flarger vofid voflume), mfid-to-hfigh vaflues for *Sal* (flong waveflength), and flow *Smr* vaflues (hfigh surface reflfiefs), on both scafles are aflfl consfistent wfith the expectatfions of pflant ffiber wear on bone (Stone, 2013). Due to fits rfigfid structure, the materfiafl does not penetrate and aflter aflfl the flevefls of the bone mficrotopography resufltfing fin rougher and flonger waveflength surfaces wfith hfigher surface reflfiefs compared to the other materfiafls, especfiaflfly skfin. Further, bark tends to produce sharper and steeper peaks of the bone mficrotopography because fit fis fless pflfiabfle than anfimafl skfin.

The resuflts of thfis study are comparabfle to those of a prevfious study that used a controflfled, mechanfized setup to create wear on bones and generate quantfitatfive data (Martfisfius et afl., 2018). The prevfious study aflso used the same confocafl dfisc-scannfing mficroscope equfipped wfith one of the flenses used fin the current study (20x). Both experfiments used sfimfiflar perfishabfle materfiafls on herbfivore rfibs, though some of the anfimafl materfiafls derfived from dfifferent taxa. A fresh *Cervus elaphus* skfin and *Bos taurus* rfibs were used fin the prevfious study (Martfisfius et afl., 2018), whfifle a semfi-fresh *Capra hfircus* skfin and *Cervus elaphus* rfibs were utfiflfized here. The other perfishabfle materfiafls (fleather and bark) were the same. The mafin dfifference fin the experfimentafl setup was the means of creatfing wear on the bones: a hfighfly controflfled mechanfized movement versus human motfion wfith added varfiabfiflfity used here. Further, the prevfious study fincfluded a wfider range of measurements wfith many scans of mfinfimaflfly worn areas, whfifle thfis study focused on the regfions compfletefly modfiffied through use. When comparfing the surface texture parameters (*Sa*, *Sal*, and *Spc* are common between studfies) of the most worn surfaces (non-modefled observatfions fin Martfisfius et afl. (2018)) and the reflatfionshfip of the materfiafl types worn between the two studfies at 20x, sfimfiflarfitfies are evfident. In both cases fresh- and semfi-fresh-skfin-worn surfaces had the flowest *Sa* vaflues (fless rough), bones used on fleather had the hfighest *Sal* vaflues (flow frequency), and those used wfith bark had the hfigher *Spc* vaflues (sharper peaks). Whfifle the parameter dfistrfibutfions of the three materfiafl types overflap to some degree, the consfistent trend between the studfies findficates that they accuratefly refflect the materfiafl propertfies of the contact materfiafls and thefir finteractfion wfith bone on the scafle observed, regardfless of the method of experfimentatfion. Thfis provfides a dfirect flfink and support for conductfing mufltfi-generatfion experfimentafl studfies (Marrefiros et afl., 2020).

5. Conclusion

The resuflts of thfis study findficate that 3D quantfitatfive mficroscopy fis usefufl for assessfing experfimentaflfly produced perfishabfle materfiafl wear on bone toofls. Moreover, evafluatfion at mufltfipfle scafles provfides addfitfionafl key finsfights finto processes that occur when bone comes finto repeated contact wfith dfifferent perfishabfle materfiafls. These quantfitatfive resuflts are compflementary to quaflfitatfive mficrowear observatfions on bone toofls. Both methods shoufld be combfined for a more compflete understandfing of the processes that occur when bone toofls are used wfith varfious materfiafls. The method presented here has the potentfiafl to aflflow researchers to access finformatfion about materfiafls that have not been preserved fin archaeoflogficafl deposfits. Stfiflfl, a sufite of bone modfifficatfions from varfious processes (e.g., manufacture, use, and post-deposfitfionafl traces) may confound dfirect appflficatfions of 3D mficroscopy to archaeoflogficafl materfiafl. Therefore, further foundatfionafl studfies are essentfiafl before thfis methodoflogy can reflfiabfly be used as an anaflytficafl toofl. At the most basfic flevefl, future studfies coufld focus on the finherent varfiatfion found fin bone fitseflf (e.g., dfifferences due to taxa, skefletafl eflement, bone state). Further, artfifacts rarefly preserve one type of surface aflteratfion, so future studfies coufld focus on assessfing bone toofls wfith dfifferfing manufacturfing traces or mufltfipurpose toofls wfith a varfiety of mficrowear traces. Taphonomfic modfifficatfions may be the bfiggest obstacfle to overcome before archaeoflogficafl materfiafl can be accuratefly assessed usfing 3D mficroscopy, sfince post-deposfitfionafl processes and thefir effects vary dependfing on myrfiad factors. Combfinfing an approach flfike the one presented here wfith sfimfiflar methods desfigned for assessfing taphonomfic aflteratfions wfiflfl be essentfiafl for estabflfishfing 3D mficroscopy as a reflfiabfle functfionafl method for evafluatfing ancfient bones. Once quantfitatfive, reproducfibfle methods are estabflfished, they can heflp to reconstruct those ephemerafl fitems and fintangfibfle processes from the archaeoflogficafl record, provfidfing finsfight finto unknown—but not unknowabfle—components of ancfient socfiafl and behavfiorafl patterns.

CRedfiT authorshfip contrfibutfion statement

Naomi L. Martisius: Conceptuaflfizatfion, Methodoflogy, Formafl anaflysfis, Investfigatfion, Data curatfion, Wrfitfing - orfigfinafl draft.

Declaration of Competing Interest

The authors decflare that they have no known competfing ffinancfiafl finterests or personafl reflatfionshfips that coufld have appeared to finffluence the work reported fin thfis paper.

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Appendix A. Supplementary data

Suppflementary data to thfis artficfle can be found onflfine at https://dofi.org/10.1016/j.jasrep.2022.103634.

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