Science
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Science **325**, 148 (2009);
DOI: 10.1126/science.1167531

Science (print ISSN 0036-8075; online ISSN 1095-9203) is published weekly, except the last week in December, by the American Association for the Advancement of Science, 1200 New York Avenue NW, Washington, DC 20005.

Comment on "DNA from Pre-Clovis Human Coprolites in Oregon, North America"

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Gilbert *et al*. (Reports, 9 May 2008, p. 786) presented DNA analysis of coprolites recovered from an Oregon cave as evidence for a human presence in North America before the Clovis culture. Results of our micromorphological and Fourier transform infrared spectroscopy analyses of one of the reported coprolites are difficult to reconcile with the DNA results identifying the coprolite as human.

Gilbert *et al*. (*1*) report the recovery, dating, and DNA analysis of coprolites from Paisley Cave, Oregon, which they ascribe to a human presence in North America about 1000 years before the Clovis culture. In 2007, the authors of the study asked us to perform micromorphological and Fourier transform infrared (FTIR) spectroscopy analyses on one of the coprolite specimens (1374-5/5D-31-2), but our results were not included in the published report (*1*). Here, we present the results of our 2007 analyses, as well as analysis of a second subsample of specimen 1374-5/5D-31-2, supplied by Gilbert *et al*. in 2009.

The subsamples we examined in 2007 and 2009 were described as follows (*2*): This specimen was found on the edge of the "bone pit" that included camel, horse, and sheep bones, a horse hoof, and other items. It is 27 mm long by 25 mm wide by 15 mm thick and appears to have an exterior surface on one side. Constituents include vegetation, feathers or fibers of some kind, small bones, and some dirt. Sample preparation, manufacture of thin sections, and micromorphological examination were carried out using protocols established in (*3*) and the supporting online material. The use of micromorphology (the study of intact sediments, soils, and materials) was a natural analytical procedure because it uses intact pieces of sample, which preserves the integrity and initial geometric arrangement of all of the components within the sample (*3*).

Microscopic evaluation of the 2007 subsample (Fig. 1) revealed the predominance of fibrous, elongated vegetative fragments, including some phytoliths. Its external morphology resembled that of herbivore dung pellets, including the typical stained outer surface (*4*, *5*). Internally, the coarse character of the plant fibers, as observed in reference thin sections of herbivore dung (e.g., camel, cattle, goat, mouflon sheep, and sheep) and as reported from the dung of browsers (*6*, *7*), is also consistent with that of herbivores. The FTIR analysis shows the presence of silicates (in the 450 to 1100 cm^{-1} region) and organic material (in the 1300 to 4000 cm^{-1} region), which are compatible with decayed organic compounds; no minerogenic phosphate was noted (Fig. 2). If phosphate is present, it is likely to be in a dominantly organic form and in low quantities (0.565% P), as is typical of dry farmyard manure [table 11.1 in (*8*)]. Examples of human coprolites and cess (mineralized sewage) are phosphate rich, containing 8 to 14% P and mean 6.57% P, respectively (*9*).

The 2009 coprolite subsample was much smaller than the first, and it appeared as a layered crust with mineral inclusions; it also contained plant and feather fragments (figs. S1 and S2). The micromorphological analysis is again

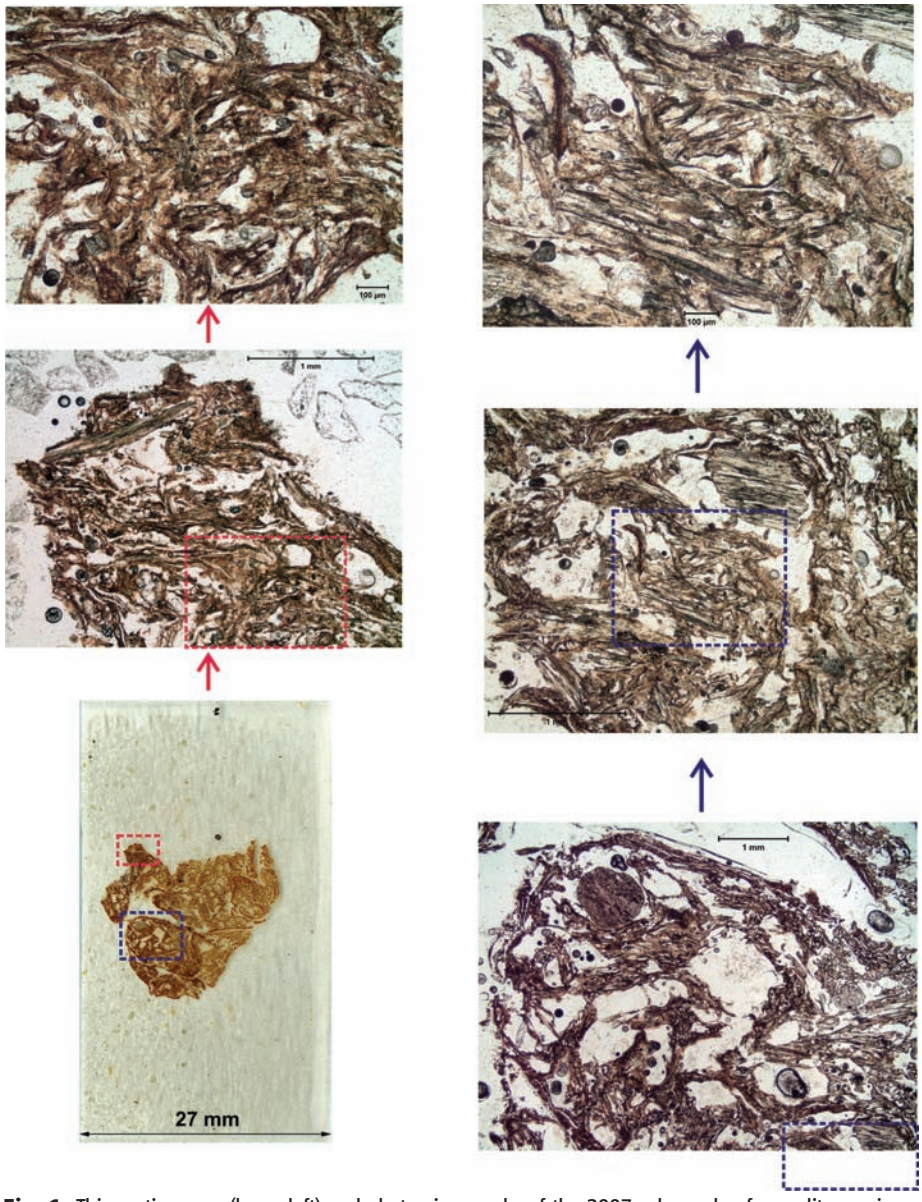

Fig. 1. Thin-section scan (lower left) and photomicrographs of the 2007 subsample of coprolite specimen 1374-5/5D-31-2 from Paisley Cave, Oregon; detailed views of different areas (rectangles) illustrate the fibrous nature of the vegetal material within the coprolite and are indicative of a herbivore origin. Contrast these views with modern camel dung and fossil hyena coprolite in fig. S4.

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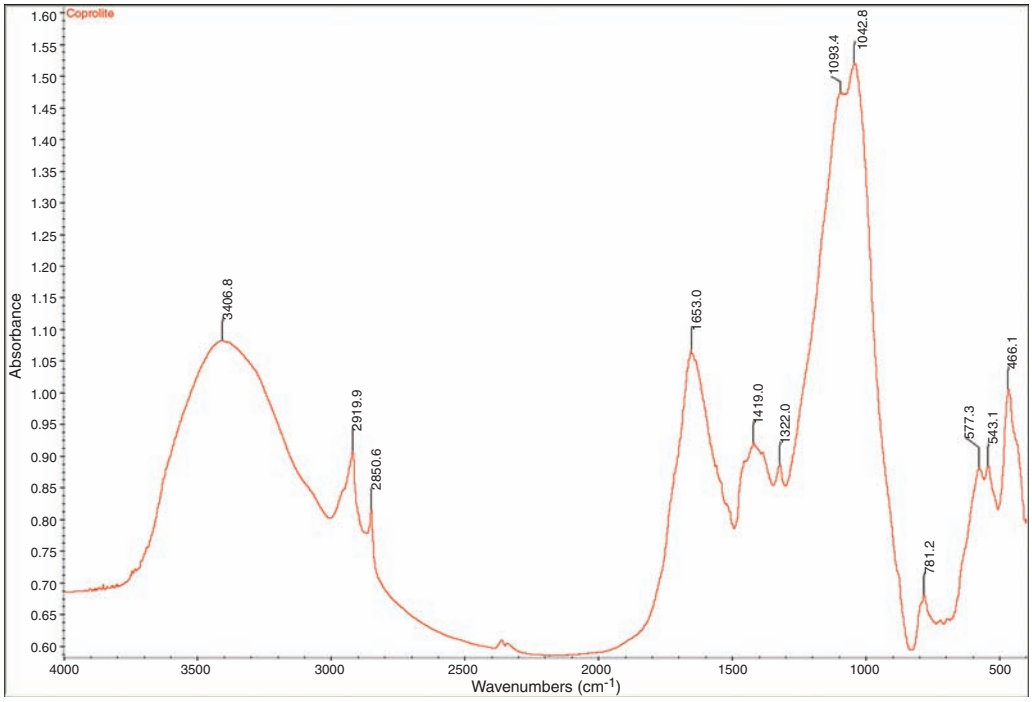

Fig. 2. Representative FTIR pattern of the 2007 subsample of coprolite specimen 1374-5/5D-31-2, which shows absorption characteristic of silicates such as feldspars, opal, quartz, and clay minerals at 465, 542, 574, 636, 695, 780, 790, 1040, and 1095 cm^{-1} and organic matter at 1320 and 1650 cm^{-1} (O-H stretching) and at 2920 and 2850 cm^{-1} (C-H stretching).

dominated by organic material (fig. S1). Very long stained plant fibers occur as an open layered structure, with occasional intercalated mineral grains (e.g., quartz). The organic matter groundmass is impregnated with micritic calcite. Overall, the micromorphological findings are compatible with those of the subsample analyzed in 2007. FTIR analyses of the bulk powdered portion and the area examined in thin section again revealed no phosphate minerals but confirmed the presence of gypsum, organic matter, quartz, clay minerals (kaolinite), and calcite (fig. S2). FTIR analysis of the feather (fig. S2) showed that the keratin is very well preserved, indicating that it is likely to be an external inclusion and did not pass through the digestive tract of a carnivore.

Interestingly, no absorption of carbonate hydroxyl apatite was present in either sample (although calcite minerals had formed in the second sample), nor was any typically yellowish amorphous cementing material observed in either thin section that could suggest a human or carnivore origin (fig. S4) [figure 3 in (*10*), figure IVa in (*3*)]. All carnivores and many omnivores, such as humans, produce calcium phosphate–rich coprolites (e.g., 18 to 34% Ca and 8 to 14% P) [appendix 1 in (*9*)], which in thin sections occur as yellowish isotropic domains that typically autofluoresce under blue light excitation (*11*). Although it is true that poor human diets can include large amounts of fiber (*12*), and large amounts of plant material such as seed remains have been found in the mummified intestines of mummies, this plant material may often still contain cellulose, which is birefringent under crossed polarized light. Humans are poorly equipped to break down cellulose, unlike herbivores; the organic material in the Paisley coprolite is mainly humified and nonbirefringent.

In sum, the subsamples of coprolite specimen 1374-5/5D-31-2 that we examined do not resemble previously described human or carnivore coprolites. The overwhelming abundance of vegetal remains (generally elongated), associated phytoliths, and lack of phosphate points to the specimen being from an herbivore (fig. S4) [figures 2 to 4 in (*4*); (*5*)]. The 2007 subsample resembles fragments of dung pellets, whereas the 2009 subsample has the layered character of trampled dung found where herbivores gather or are in relatively confined spaces (*5*, *13*). Both sets of micromorphological and FTIR findings are incompatible with the coprolite specimen 1374-5/5D-31-2 being of human origin, and thus are inconsistent with the DNA results presented in (*1*).

References and Notes

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14. The infrared spectroscopy data furnished here was made possible by a grant from the National Science Foundation to P. Goldberg (BCS 0551927) and the support by a Marie Curie International Fellowship within the 6th European Community Framework Programme to F. Berna (MOIFCT-2006-041053).

Supporting Online Material

www.sciencemag.org/cgi/content/full/325/5937/148-c/DC1
Materials and Methods
Figs. S1 to S4
References

22 October 2008; accepted 16 June 2009
10.1126/science.1167531