Supporting Information for:

Preparation of artificial antibodies and development of an antibody-

based indirect ELISA for the detection of ancient wool

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Fig.S1 SDS-PAGE analysis of keratin extracted from wool. Lane 1: Molecular weight marker; lane 2: keratin after heat treatment at 100 °C for 10 min; lane 3: keratin without heat treatment.

The molecular weight distribution (MWD) of the resultant keratin was determined by SDS-PAGE under reducing conditions. Depending upon the molar mass and sulphur content, four fractions of proteins can be extracted from wool, namely the low sulphur fraction (LSF, Mw: 45-60 kDa), the high (HSF, Mw: 14-28 kDa) and ultrahigh sulphur fraction (USF, Mw: 37 kDa), and the high Gly/Tyr fraction (HGT, Mw: 9-13 kDa).¹ As shown in Fig. S1, two bands that match the distribution characteristics of the typical keratin bands were observed, including the HSF chain at approximately 28 kDa and the HGT at approximately 12 kDa, indicating that the resultant keratins were mainly the HSF and HGT fractions.



Fig.S2 FTIR spectra of wool and keratin.

The internal structure and chemical composition of the resultant keratin was characterized via FTIR. As shown in Fig. S2, both the resultant keratin and wool show the characteristic absorption peak of polypeptide at 3080 cm⁻¹. Also, the sample spectra show the characteristic peaks of amide bonds, specifically a sharp peak at 1654 cm⁻¹ (amide I band), a peak at 1543 cm⁻¹ (amide II band) and a peak at 1240 cm⁻¹ (amide III band). In particular, a peak at 1045 cm⁻¹ appears in the spectrum of resultant keratin, which is assigned to the characteristic peak of cysteinesulfenate produced by oxidation of disulfide bond.

References

(1) Simpson, W. S.; Crawshaw, G. H. *Wool: Science and technology*; Woodhead Publishing Limited: Cambridge, 2002.