Winterthur/University of Delaware
Program in Art Conservation

Investigation of Commercially Available Rabbit Skin Glues Using MALDI-TOF Mass Spectrometry

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ABSTRACT: Rabbit skin glue is often indicated as a preferred binder for the preparatory layers in traditional water gilding and as a treatment material in gilded objects conservation and restoration, but previous studies have suggested that references to this material are mostly absent from pre-twentieth-century recipes, treatises, and literature and that some modern preparations sold as 'rabbit skin glue' may in fact be generic animal collagen glues. Thirteen commercially available or commercially purchased products sold as 'rabbit skin glue' were analyzed by peptide mass fingerprinting (PMF) with matrix-assisted LASER desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) in order to characterize their proteins and suggest a species- or family-specific animal source. Five of the products analyzed had protein fingerprints consistent with cattle, with no indication of any protein mixture; three of the products analyzed had protein fingerprints consistent with rabbit, with no indication of a mixture; and five of the products had a protein fingerprint consistent with a mixture of proteins, either cattle protein and rabbit protein, or cattle protein and another undetermined animal protein. This study provides scientific results that prove that some of these materials' compositions are not consistent with their advertised or implied compositions; this study can therefore be referenced by future researchers or users of these products to inform their work.

Keywords: Rabbit skin glue; rabbitskin glue; hide glue; animal protein glue; size; collagen; PMF; MALDI-TOF MS; proteomics; gilding; gesso ground; bole; traditional materials; artist's materials.

1. Introduction

Rabbit skin glue is often indicated as a preferred binder for the preparatory layers in traditional water gilding and as a treatment material in gilded objects conservation and restoration (Rivers and Umney 2013, 647-648, 773-774; von Reventlow 1991, 269). Seventeenth- and eighteenth-century British gilding treatises do not mention rabbit skin glue and instead recommend a glue made from scraps of parchment or glover's leather as a binder for gilding preparations (Stalker and Parker 1688, 57; Campbell 1747, 107-108; Dossie 1764, 433-434), and the eighteenth-century French author Jean-Felix Watin also recommends parchment glue for gilding in his 1772 treatise (Watin 1772, 151). A 2005 survey of sixteenth- to nineteenth-century European recipes for painting grounds found no mention of rabbit skin glue in the recipes it examined (Witlox and Carlyle 2005).[1] Rabbit skin glue is mentioned as early as 1876 in the anonymous publication *The Gilder's Manual,* as a French gilder's size (Anonymous 1876), and it seems that the use of this glue may have emerged in France and Germany using byproducts of the hat industry, which required rabbit hair to make felt (Wehlte 1975, 372).

There is anecdotal evidence that modern "rabbit skin" glues may often be made from generic animal skins (Carlyle 2012, 35). The Museum of Fine Arts Boston's Conservation and Art Materials Encyclopedia Online defines rabbit skin glue as, "a strong, water-soluble adhesive obtained from the hydrolyzed products of the skin of rabbits," but this definition does not address the historical use of this

material or the composition of currently available commercial products (Museum of Fine Arts Boston, 2016). A 2011 study used peptide mass fingerprinting (PMF) to characterize several samples of rabbit skin glues. The study found that some samples were consistent with rabbit peptides and that some were consistent with a bovine peptides, however the commercial provenances of the samples found to be consistent with bovine peptides were intentionally withheld by or were unknown to the authors (Dallongeville, Koperska, Garnier, Reille-Taillefert, Rolando, and Tokarski 2011).[2]

Proteomics techniques like MALDI-TOF MS provide tools that are capable of discriminating between proteins with great specificity, often to the family or species level (Buckley, Collins, Thomas-Oates, and Wilson 2009). MALDI-TOF MS allows for the detection and characterization of large synthetic polymers and biopolymers (like collagen) which are difficult to ionize and desorb into the gas phase for mass spectrometry by traditional techniques (e.g. electron impact) without being destroyed in the process. Samples are digested by a protein (e.g. trypsin) and combined with a matrix (e.g. α-cyano-4-hydroxycinnamic acid (CHCA)), a small organic molecule containing a conjugated pi system which absorbs UV energy from the laser when the sample is ionized during mass spectrometry, preserving its integrity (Anoka-Ramsey Community College 2016). This technique has gained popularity in cultural heritage research because sample-size requirements are small, sample preparation is relatively straightforward, the technique can discriminate between proteins more specifically than other common analytical techniques like gas chromatography-mass spectrometry (GC-MS), and aged or degraded proteins may be accurately characterized (Buckley et al. 2009; van der Werf, Calvano, Palmisano, and Sabbatini 2012). MALDI-TOF has been used to study gilded objects and gilders' materials (Dallongeville et al. 2011; Kučková, Schultz, Veiga, Murta, and Crina Anca Sandu 2015). While Dallongeville et al.'s previously-mentioned study effectively demonstrates the use of this technique for the characterization of the source species for various protein glues, it has limited utility for artists, craftspeople, conservators, and restorers seeking to make informed choices about rabbit skin glues in practical use because most of the rabbit skin glues studied are either unidentified or are not commonly available.

2. Experimental

Eight products marketed as "genuine rabbit skin glue" or "rabbit skin glue" and/ or explicitly claiming in their technical data sheets or marketing materials to be made from rabbits were purchased online for the purpose of family- or species-specific protein characterization using PMF with MALDI-TOF MS. Four products bought as "rabbit skin glue" were also obtained from Behrooz Salimnejad, Senior Conservator of Furniture at the Philadelphia Museum of Art, for analysis in this study. One unlabeled product found in Winterthur Museum's Furniture Conservation Lab was also tested. The following products were tested: *Utrecht Genuine Rabbit Skin Glue and Size* (RSG1), *Kremer Pigments Rabbit Skin Glue Cubes* (RSG2), *Williamsburg Genuine Rabbit Skin Glue* (RSG3), and *Cornelissen Genuine Rabbit Skin Glue* (RSG4), *Rublev/ Natural Pigments Genuine Rabbit Skin Glue* (RSG5), *Greg Dorrance Rabbit Skin Glue, German Granular* (RSG 6), *Woodfinishing Enterprises Imported Rabbit Skin Glue* (RSG7), *A.S. Handover Rabbit Skin Glue* (RSG8), *Sepp Leaf Rabbit Skin Glue: ~10 years old (obtained from Behrooz Salimnejad)* (RSG9), *Sepp Leaf Rabbit Skin Glue: ~3 years old (obtained from Behrooz Salimnejad)* (RSG10), *Iranian Rabbit Skin Glue (obtained from Behrooz Salimnejad)* (RSG11), *German Rabbit Skin Glue (obtained from Behrooz Salimnejad)* (RSG12), *Unidentified Glue (obtained from Winterthur Furniture Lab)* (RSG13). Excerpted technical data sheets and/ or marketing materials for each product analyzed, if available, are provided in Appendix 1.

2.1 Methodology

The experimental procedure was provided by Dr. Dan Kirby and is detailed in Appendix 2. The following procedure was repeated for each sample:

Sample preparation:
1. Sample placed in 600 µL Eppendorf tube (V-Vial) using a clean scalpel and solubilized in 30 µL trifluoroethanol, 30 µL 50 mM ammonium bicarbonate (AMBI) at 60 ºC for 45 min with intermittent vortexing/agitation;
2. Digested with 8 µL trypsin (0.02 ug/µL in 50 mM AMBI) at 37 ºC overnight.

MALDI-TOF analysis:
1. A-cyano-4-hydroxycinnamic acid (CHCA) matrix prepared as a saturated solution in 40% (v- v) acetonitrile (ACN) – 0.1% trifluoroacetic acid (TFA);
2. Samples (4 µL) mixed with matrix (20 µL) in a separate tube and spotted on the MALDI plate;
3. Peptide standards were used as external calibrants;
4. A Bruker MicroFlex MALDI-TOF at the University of Delaware Mass Spectrometry facility was used to collect positive ion spectra from 800 – 3800Da with mass accuracy of ± 0.1 Da.

Data analysis:
1. The resulting spectrum was exported as a .txt file and analyzed using mMass open source mass spectrometry software.
2. Results were compared to PMF markers from known reference samples to identify collagen-based materials.

3. Results

Three of the products tested were found to have peptide mass fingerprints clearly consistent with rabbit collagen with no indication of a mixture (RSG 7, RSG11, and RSG 13); five of the products tested were found to have peptide mass fingerprints clearly consistent with cattle/ bison collagen with no indication of a mixture (RSG 2, RSG 3, RSG4, RSG5, and RSG6); one of the products tested had a peptide mass fingerprint clearly consistent with a mixture of rabbit collagen and cattle/ bison collagen (RSG9); four products tested had peptide mass fingerprints consistent with a mixture of proteins, either cattle protein and rabbit protein, or cattle protein and another undetermined animal protein (RSG1, RSG8, RSG10, RSG12) (Table 1). Results were interpreted with guidance from Catherine Matsen, Dr. Kate Duffy, and Dr. Dan Kirby. MALDI-TOF MS data and reference PMF markers are provided in Appendix 3.

Table 1. Experiment results

Product tested	Comments
Utrecht Genuine Rabbit Skin Glue & Size – *granules* (RSG1)	Presence of two (B) markers and two (F) markers indicates a mixture. No (C) markers observed. Likely contains cattle and others to be determined. Contains three rabbit markers (A, B, F), however most expected rabbit markers are not observed.
Kremer Pigments Rabbit Skin Glue – *cubes* (RSG2)	Shares all markers with cattle/ bison except for (G) which is not needed for identification. There is no indication of a mixture.
Williamsburg Rabbit Skin Glue – *granules* (RSG3)	Shares all markers with cattle/ bison except for (G) which is not needed for identification. There is no indication of a mixture.
L. Cornelissen & Son Genuine Rabbit Skin Glue – *granules* (RSG4)	Shares all markers with cattle/ bison except for (G) which is not needed for identification. There is no indication of a mixture.

Rublev/ Natural Pigments Genuine Rabbit Skin Glue – *granules* (RSG5)	Shares all markers with cattle/ bison. There is no indication of a mixture.
Greg Dorrance Rabbit Skin Glue, German Granular – *granules* (RSG6)	Shares all markers with cattle/ bison. There is no indication of a mixture.
Woodfinishing Enterprises Imported Rabbitskin Glue – *sheets* (RSG7)	Shares all markers with rabbit except for (C) and (G) which are not needed for identification. There is no indication of a mixture.
A.S. Handover Rabbit Skin Glue – *granules* (RSG8)	Shares all markers with cattle/ bison except for (G), which is not necessary for identification. There are two (F) markers, indicating a mixture. (F) marker 2883 is shared by many species, including rabbit. No other rabbit markers present.
Sepp Leaf Rabbit Skin Glue: ~10 years old – *granules* (RSG9)	Shares all markers with cattle/ bison except for (G), which is not needed for identification. Shares all markers with rabbit except for (G), which is not needed for identification. Presence of two markers for all markers except for (G) indicates a mixture of rabbit and cattle/ bison. (D) markers for rabbit and for cattle/ bison are very close to each other and are difficult to resolve with certainty.
Sepp Leaf Rabbit Skin Glue: ~3 years old – *cubes* (RSG10)	Shares all markers with cattle/ bison except for (C) and (G), which are not needed for identification. There are two (F) markers, indicating a mixture. (F) marker 2883 is shared by many species, including rabbit. Could have (D) marker for rabbit; (D) markers for rabbit and for cattle/ bison are very close to each other and are difficult to resolve with certainty.
Iranian Rabbit Skin Glue – *sheets* (RSG11)	Shares all markers with rabbit except for (C) and (G) which are not needed for identification. There is no indication of a mixture.
German Rabbit Skin Glue – *cubes* (RSG12)	Shares all markers with cattle/ bison except for (C), which is not necessary for identification. Shares (B), (F), and possibly (D) markers with rabbit. (D) markers for rabbit and for cattle/ bison are very close to each other and are difficult to resolve with certainty.
Unidentified Glue (from Winterthur) – *sheets* (RSG13)	Shares all markers with rabbit except for (C) which is not needed for identification. There is no indication of a mixture.

4. Discussion

It is important to distinguish between animal protein glues in art, craft, restoration, and conservation practices because different grades of glue and glues derived from different animals can have different properties (Schellmann 2007). The longevity of artworks may be significantly affected by the artist's choices and understanding of materials. Likewise, the effective and ethical implementation and documentation of conservation treatments depends on an informed understanding of the materials used in the object's original manufacture and those introduced later as restoration materials. This study provides scientific results that prove that some of these materials' compositions are not consistent with their advertised or implied compositions; this study can therefore be referenced by future researchers or users of these products to inform their work.

Further research may include the identification of the protein content in a wider selection of commercially available rabbit skin glues and experimental study of their physical properties, such as bloom strength (gram weight strength), in further service of demystification and informed practice.

Acknowledgments
Thanks to Catherine Matsen, Dr. Dan Kirby, Dr. Rosie Grayburn, Lara Kaplan, Sarah Towers, and Kate Acuna, who prepared samples in my absence and to Dr. Papa Nii Asare-Okai who performed MALDI-TOF MS at the University of Delaware. Thanks also to Dr. Kate Duffy and Behrooz Salimnejad at the Philadelphia Museum of Art and to Mark Anderson and Kathy Gillis at Winterthur Museum. This study was inspired in part by conversations with Dr. Joyce Hill Stoner.

Notes

1. Preparatory layers for painting, like gilding, often use an animal protein binder or size.

2. A sample from Kremer Pigments, a historic sample from the French firm of Maison Totin Frères, and a sample of unknown commercial provenance were found by Dalongeville et al. to be consistent with rabbit peptides. A sample from an unnamed commercial source and two samples of unknown commercial provenance were found to be consistent with bovine rather than rabbit peptides.

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Sources of Materials

Utrecht Genuine Rabbit Skin Glue & Size (#:08943-1001)
Blick Art Materials
PO Box 1769
Galesburg, IL 61402-1769

Kremer Rabbit Skin Glue Cubes (#63025)
Kremer Pigments
247 W 29th St C
New York, NY 10001

Williamsburg Genuine Rabbit Skin Glue (#79948)
Golden Artist Colors, Inc.
188 Bell Road
New Berlin, NY 13411-9527

Cornelissen Genuine Rabbit Skin Glue (# LC27484H)
L. Cornelissen & Son Artist’s Colourmen
105 Great Russell St,
Bloomsbury, London WC1B 3RY
United Kingdom

Rublev Genuine Rabbit Skin Glue (#510-21GRS50)
Natural Pigments
291 Shell Lane
Willits, CA 95490

Greg Dorrance Rabbit Skin Glue, German Granular (#Sep-RSG33503-1)
Greg Dorrance Co.
1063 Oakhill Ave.
Attleboro, MA 02703-7318

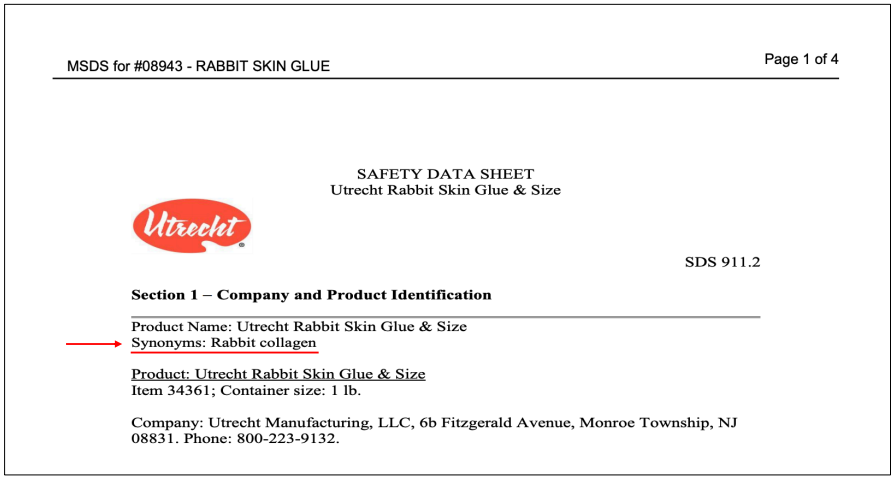
Woodfinishing Enterprises Imported Rabbitskin Glue (#75-5020-5)
WoodFinishing Enterprises
1729 North 68th Street
Wauwatosa, WI 53213

A.S. Handover Rabbit Skin Glue (#MHRSG500)
Jackson's Art Supplies
1 Farleigh Place
London, N16 7SX
United Kingdom

Appendices

Appendix 1. Excerpts from technical data sheets and/ or marketing materials for each product analyzed

Notations in red are added for clarity.



MSDS for #08943 - RABBIT SKIN GLUE Page 1 of 4

SAFETY DATA SHEET
Utrecht Rabbit Skin Glue & Size

SDS 911.2

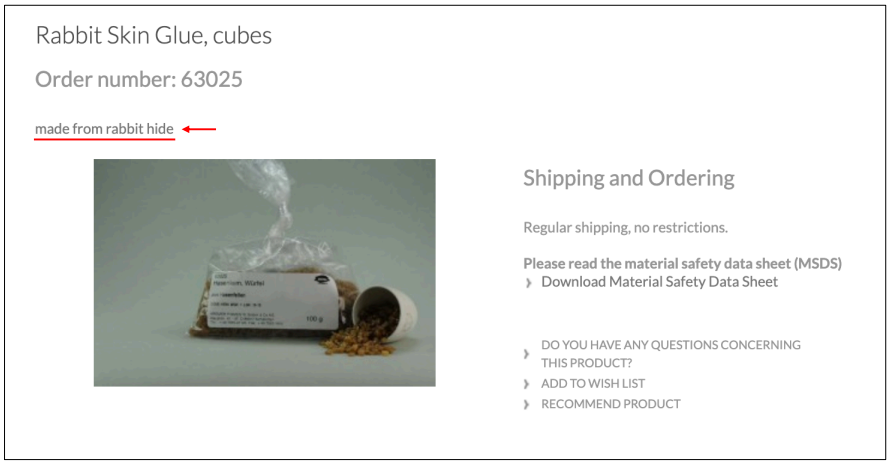
Section 1 – Company and Product Identification

Product Name: Utrecht Rabbit Skin Glue & Size
Synonyms: Rabbit collagen

Product: Utrecht Rabbit Skin Glue & Size
Item 34361; Container size: 1 lb.

Company: Utrecht Manufacturing, LLC, 6b Fitzgerald Avenue, Monroe Township, NJ 08831. Phone: 800-223-9132.

Figure 1. Excerpt from *Utrecht Genuine Rabbit Skin Glue & Size* manufacturer's safety data sheet.
https://cdn.dick-blick.com/msds/DBH_SDS_089431001.pdf (accessed 5/4/2020.



Rabbit Skin Glue, cubes

Order number: 63025

made from rabbit hide

Shipping and Ordering

Regular shipping, no restrictions.

Please read the material safety data sheet (MSDS)
Download Material Safety Data Sheet

DO YOU HAVE ANY QUESTIONS CONCERNING THIS PRODUCT?
ADD TO WISH LIST
RECOMMEND PRODUCT

Figure 2. Screenshot from Kremer Pigments website.
https://shop.kremerpigments.com/en/mediums-binders-und-glues/water-soluble-binders/natural-glues-und-agglutinants/5719/rabbit-skin-glue-cubes (accessed 5/4/2020).

TECH SHEET

Williamsburg
HANDMADE OIL COLORS

RABBIT SKIN GLUE

General Description

A genuine rabbit skin glue made from the hide and bones of rabbits. Comes as a crystalized powder. Commonly used as a size, an adhesive and a component of traditional gesso in painting.

Gel/Bloom strength: 550 grams

Figure 3. Excerpt from *Williamsburg Genuine Rabbit Skin Glue* technical data sheet. https://goldenhub.goldenpaints.com/storage/uploads/rabbit-skin-glue-tech-sheet.pdf (accessed 5/4/2020).

L. CORNELISSEN & SON
Artists' Colourmen
Suppliers of Materials for Painters, Gilders & Printmakers

Safety Data Sheet according to Regulation (EC) No. 1907/2006

Revision Date: October 2015

1) Identification of the substance/preparation and the company

Trade Name: Cornelissen Rabbit Skin Glue Powder/Granules.

Application: Artists' Size.

Manufacturer/Supplier:

L Cornelissen & Son Ltd
105 Great Russell Street
London WC1B 3RY

Tel: 020 7636 1045
Fax: 020 7636 3655

www.cornelissen.com

2) Composition/Information on ingredients

Rabbit Skin Glue is not classified as hazardous.

Figure 4. Excerpt from *L. Cornelissen & Son Genuine Rabbit Skin Glue* safety data sheet. https://www.cornelissen.com/media/attachment/file/m/s/msds_pgr_rsg_granules.pdf (accessed 5/4/2020).

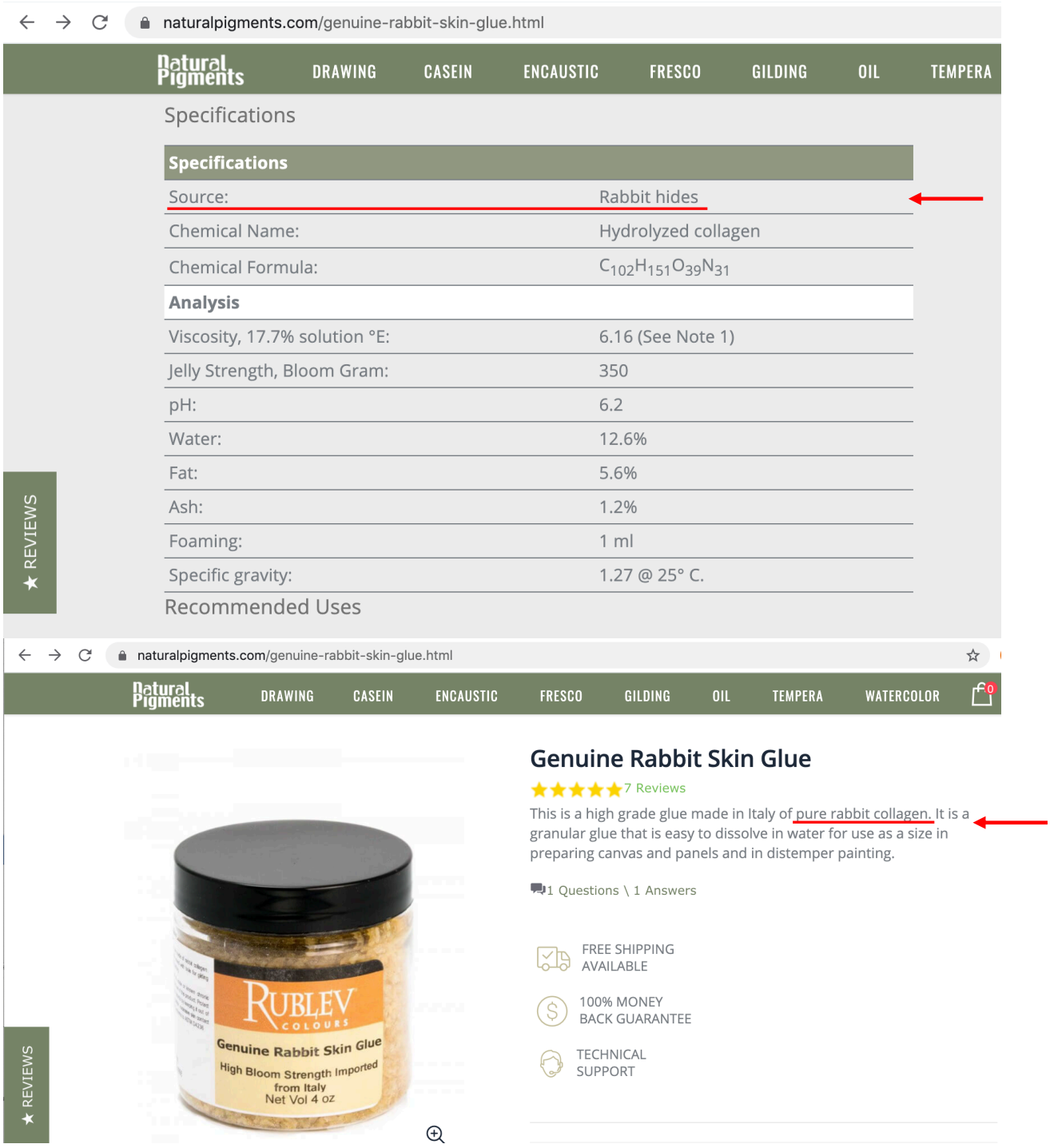


Figure 5. Screenshots from Natural Pigments website. https://www.naturalpigments.com/genuine-rabbit-skin-glue.html (accessed 7/25/2021).

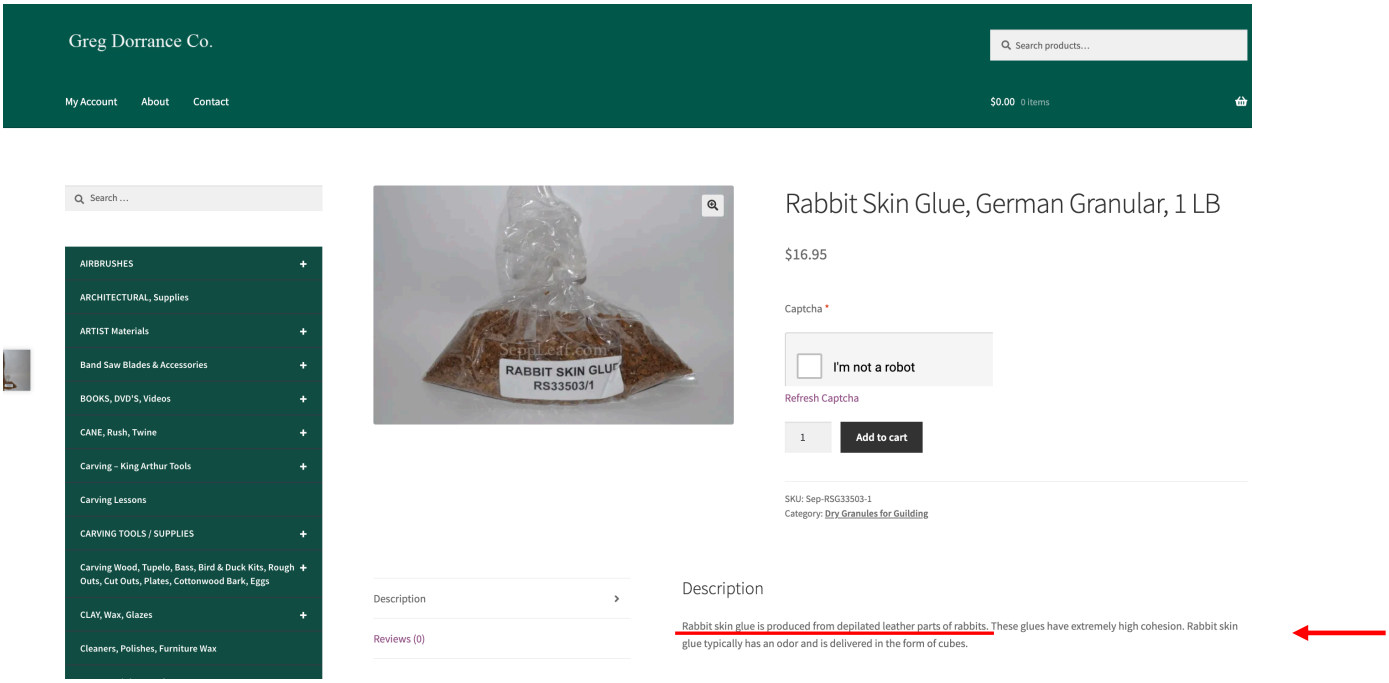


Figure 6. Screenshot from Greg Dorrance Co. website. https://gregdorrance.com/product/rabbit-skin-glue-german-granular-1-lb/ (accessed 7/25/2021).

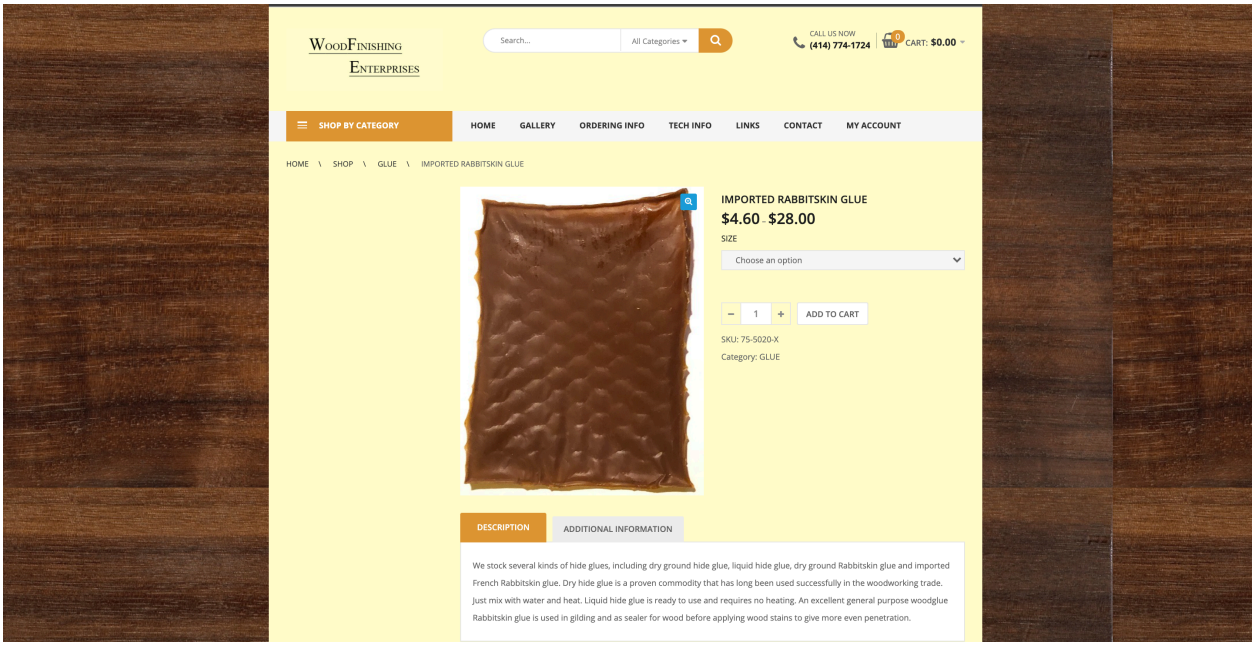


Figure 7. Screenshot from WoodFinishing Enterprises website. https://woodfinishingenterprises.com/shop/glue/imported-rabbitskin-glue/ (accessed 7/25/2021).

Figure 8. Screenshot from A.S. Handover website. https://www.handover.co.uk/handover-prepared-rabbit-skin-glue (accessed 7/25/2021).

Appendix 2. Detailed experimental procedure

Experimental procedure provided by Dr. Dan Kirby

General Considerations:
Place several grains or particles of sample in a 600 µL Eppendorf tube (V-Vial) using a clean scalpel.

Solubilization:
1. In clean hood, add 60 µL 50 mM AMBI (ammonium bicarbonate) to each sample. Vortex and spin (mini centrifuge) to solubilize the protein sample. 50 mM AMBI has a pH of approximately 8.3, which is optimal for trypsin activity. To prepare 50mM AMBI, dilute 100 mM AMBI (79 mg AMBI in 10mL HPLC grade H_2O) to 50 mM and check that pH is 8.3. Make fresh solution after 1 week to avoid bacterial growth.

2. Heat samples at 60 °C for 45 minutes. Check samples in V-vials under the microscope, if necessary, to make sure that the sample is in the liquid. If not, flick down solid on walls. The sample must be in the liquid in order for it to be solubilized.

Reduction and Alkylation:
3. Cool samples to room temperature. Add 3 µL 20 mM TCEP (tris(2-carboxyethyl) phosphine hydrochloride) in 25 mM AMBI. Heat to 37 °C for 15 min. TCEP reduces disulfide bonds in proteins to thiols aiding in solubilization and tryptic digestion. 20 mM TCEP in 25 mM AMBI: 57.3 mg TCEP in 10 mL 25 mM AMBI. Freeze 100-200 µL aliquots until needed.

4. Add 3 µL 40 mM IAA (iodoacetamide) in 25 mM AMBI. Allow to react for 15 minutes, at room temperature and protected from light. IAA end caps the thiols formed in the previous step and prevents

them from recombining. 40 mM IAA in 25 mM AMBI: 74 mg IAA in 10 mL 25 mM AMBI. Freeze 100-200 µL aliquots until needed. Protect from light.

Digestion:

6. Add 8 µL trypsin (0.02 µg/µL in 50 mM AMBI). Trypsin digests protein into predictable peptides by cutting the protein on the CC-terminal side of lysine (K) and arginine (R). Trypsin 0.02 µg/ µL 50 mM AMBI to a vial containing 20 µg lyophilized trypsin. Freese 100 µL aliquots until needed.

7. Digest at 37 °C overnight. Digestion may be essentially complete in as little as 2 hours. For convenience, digests are usually left overnight.

Sample Preparation for MALDI:

8. Prepare matrix: Saturated CHCA (α-cyano-4-hydroxycinnamic acid) in 40% (v/v) ACN (Acetonitrile) 0.1% (v/v) TFA. Add a small amount of CHCA to a glass vial, add 40% ACN 0.1% TFA, vortex intermittently for 5 minutes and allow undissolved CHCA to settle. If undissolved CHCA is present, the solution is saturated.

9. Prepare samples: Combine 20 µL matrix and 2-3 µL sample in a new 600 µL tube. Vortex and spin. Spot ~1.5 µL on the Maldi plate. Allow to air dry. Do not allow pipette tip to touvh the MALDI plate as this interferes with proper matrix crystallization. Check dried spots under the microscope to observe proper crystallization.

10. Prepare peptide standard: 3 µL standard and 20 µL matrix. Spot ~1 µL on the MALDI plate. Allow to air dry. Do not allow pipette to touch the MALDI plate.

MALDI:

11. A Bruker MicroFlex MALDI-TOF at the University of Delaware Mass Spectrometry facility was used to collect positive ion spectra from 800 - 3800Da with mass accuracy of ± 0.1 Da.

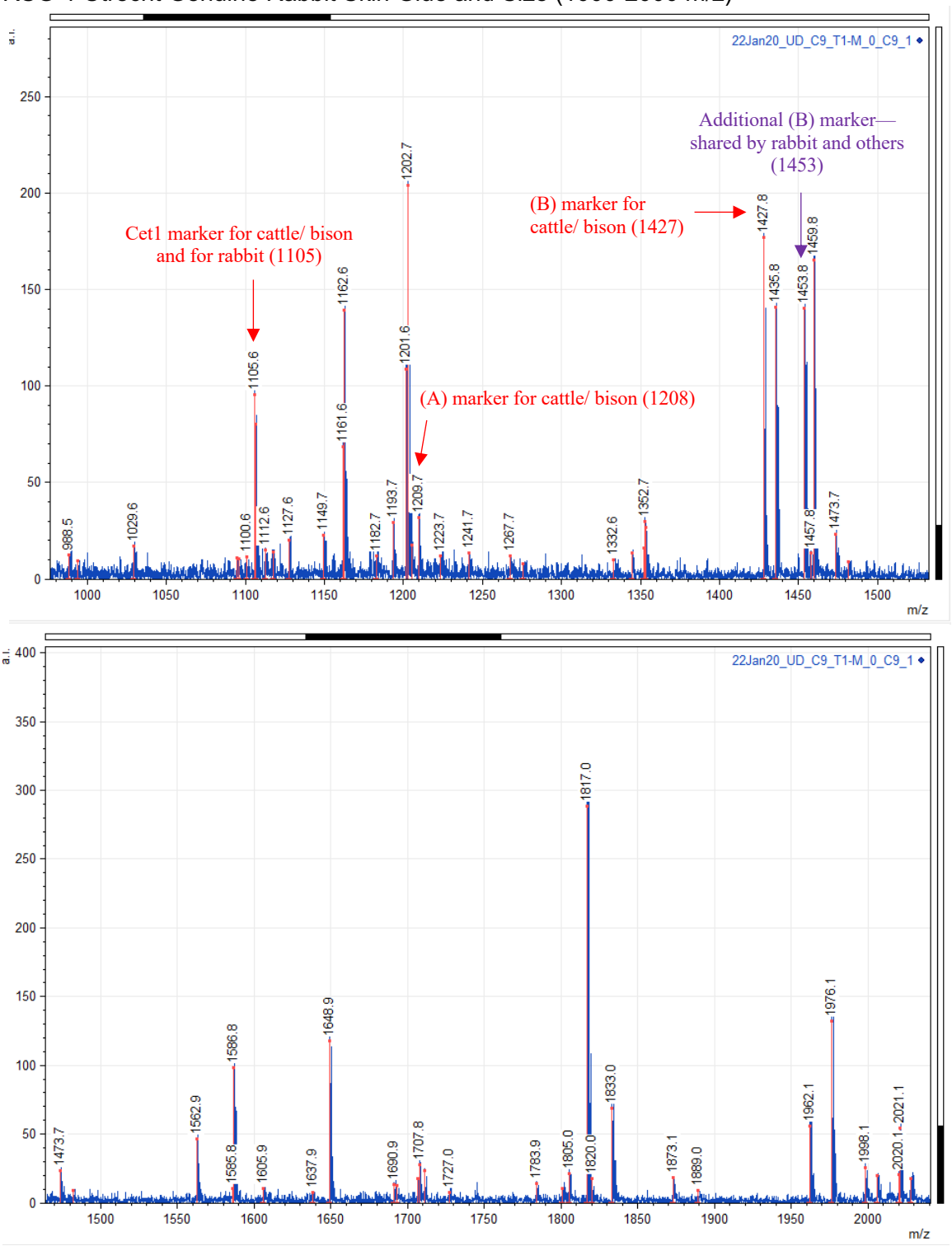
Data analysis:

12. The resulting spectrum was exported as a .txt file and analyzed using mMass open source mass spectrometry software.
13. Results were compared to PMF markers from known reference samples to identify collagen-based materials.

Appendix 3. MALDI-TOF MS spectra and PMF reference markers

Results were interpreted with guidance from Catherine Matsen, Dr. Kate Duffy, and Dr. Dan Kirby.

RSG-1 Utrecht Genuine Rabbit Skin Glue and Size (1000-2000 m/z)



RSG-1 Utrecht Genuine Rabbit Skin Glue and Size (2000-3200 m/z)

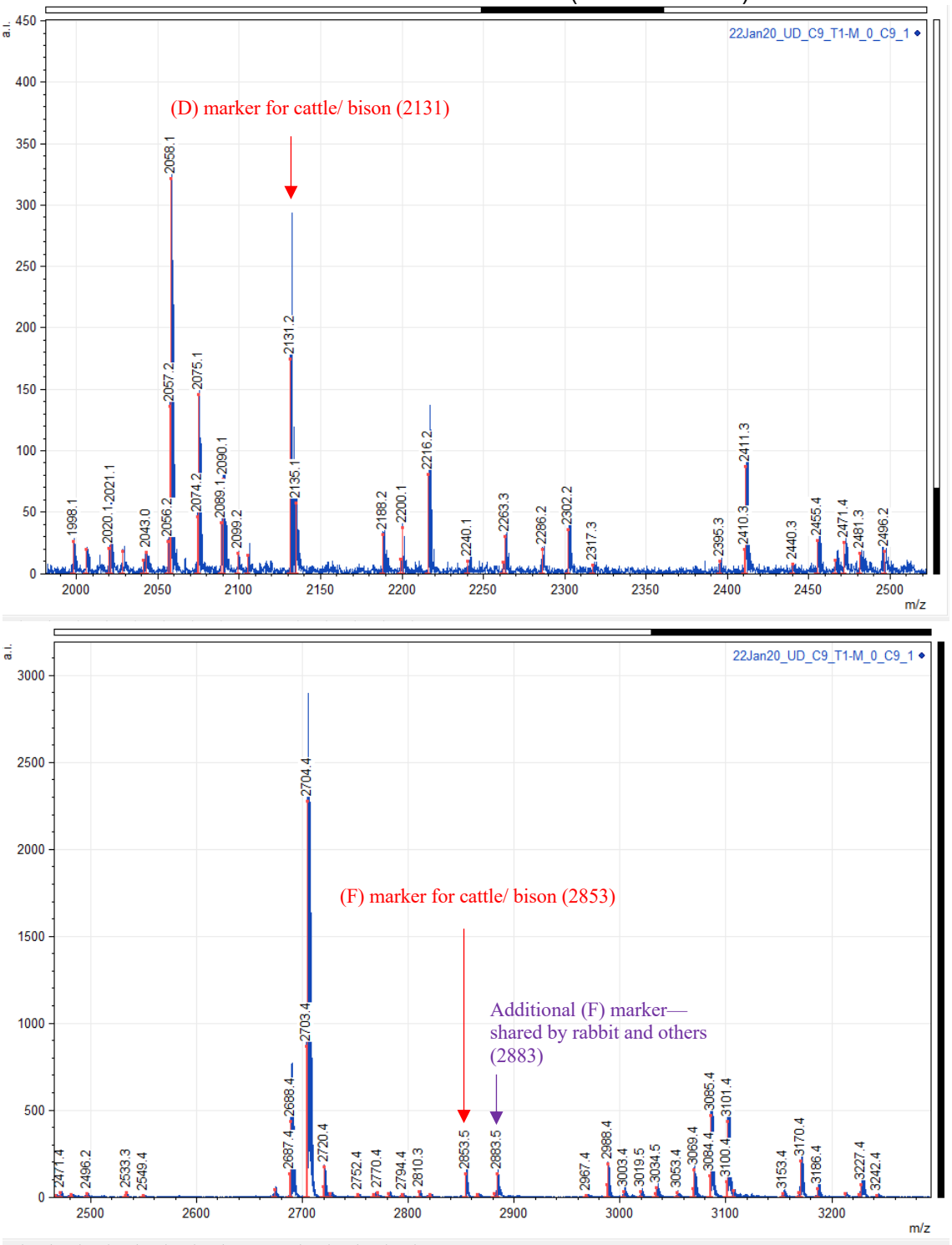
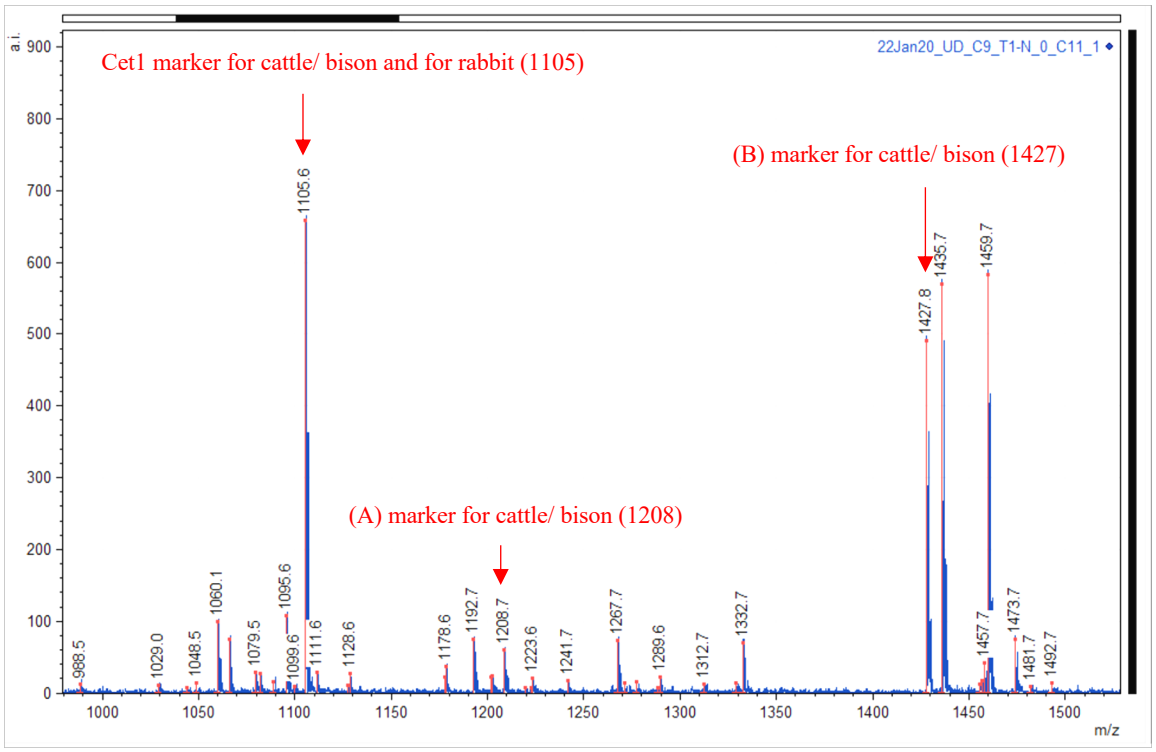
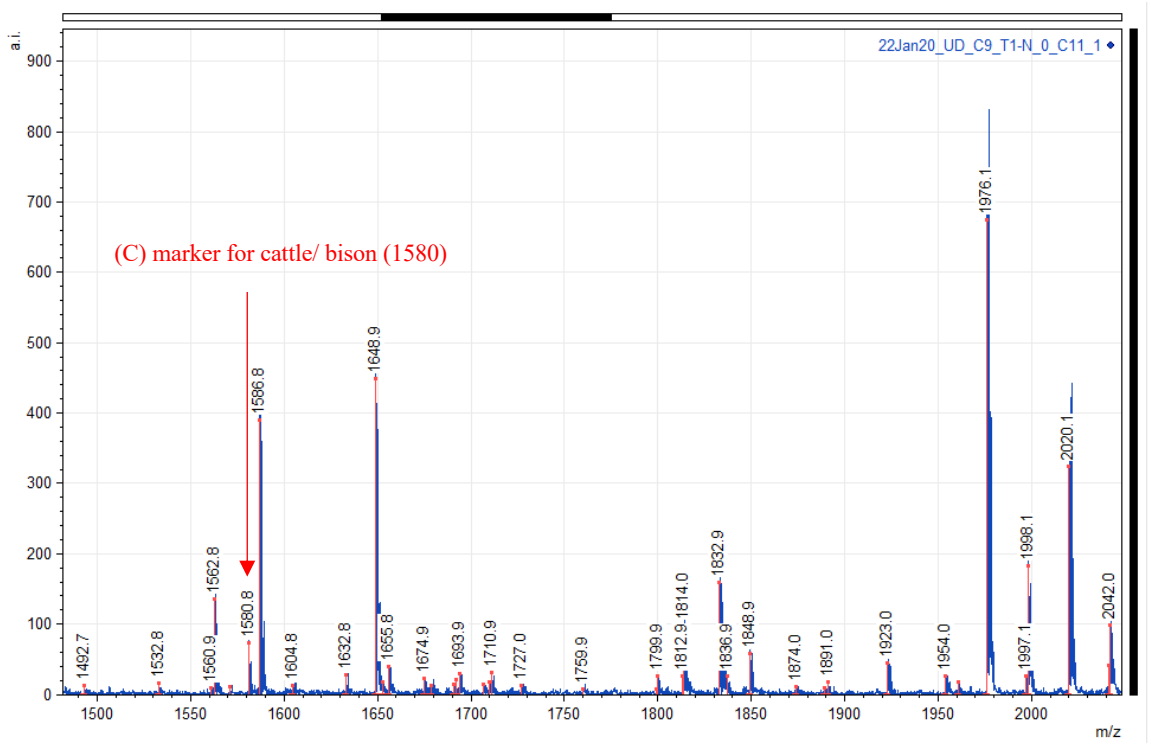


Figure 9. MALDI-TOF MS spectra for RSG1–*Utrecht Genuine Rabbit Skin Glue and Size* 1000-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Presence of two (B) markers and two (F) markers indicates a mixture. No (C) markers observed. Likely contains cattle and others to be determined. Contains three rabbit markers (A, B, F), however most expected rabbit markers are not observed.

RSG-2 Kremer Pigments Rabbit Skin Glue (1000-2000 m/z)





RSG-2 Kremer Pigments Rabbit Skin Glue (2000-3200 m/z)

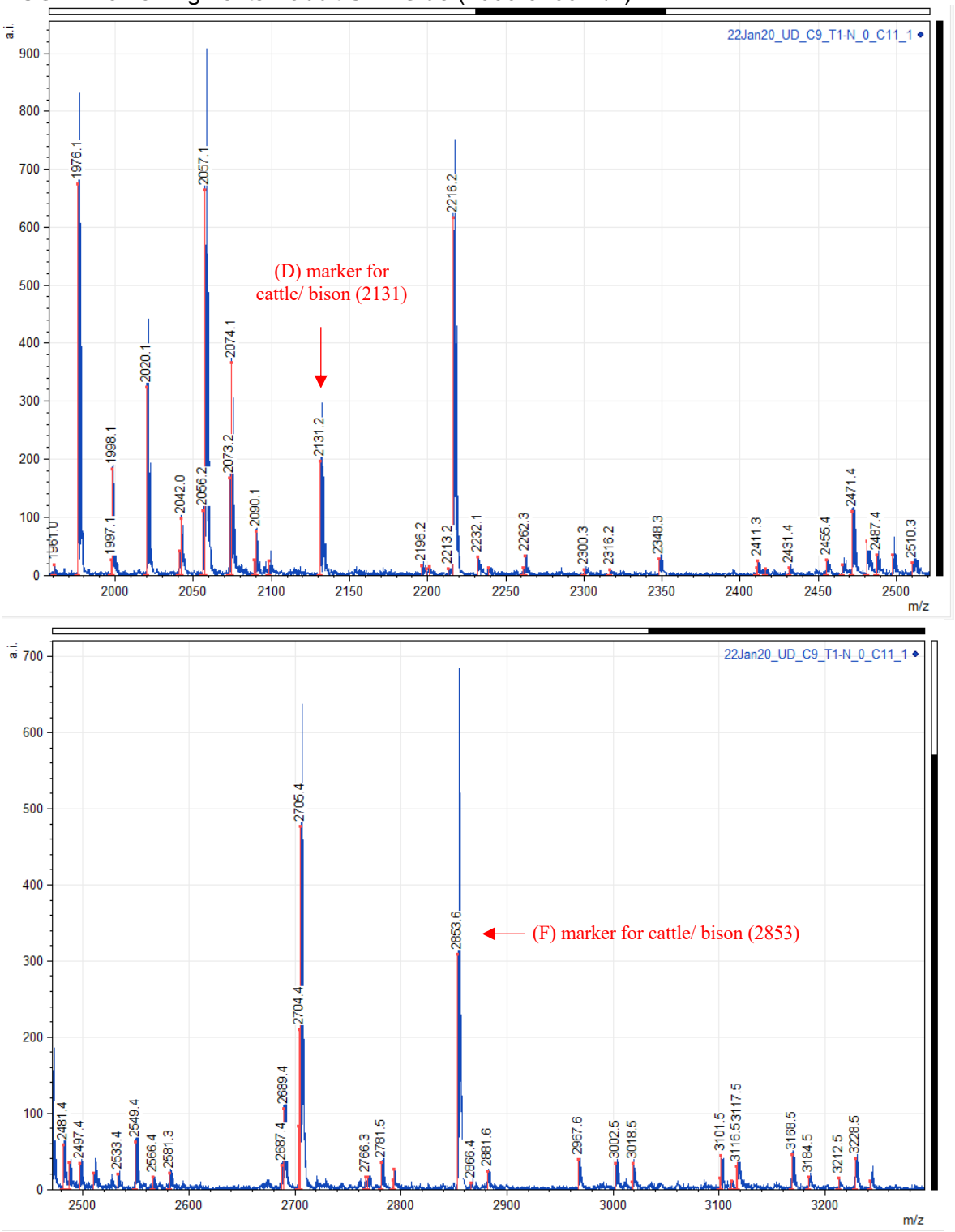
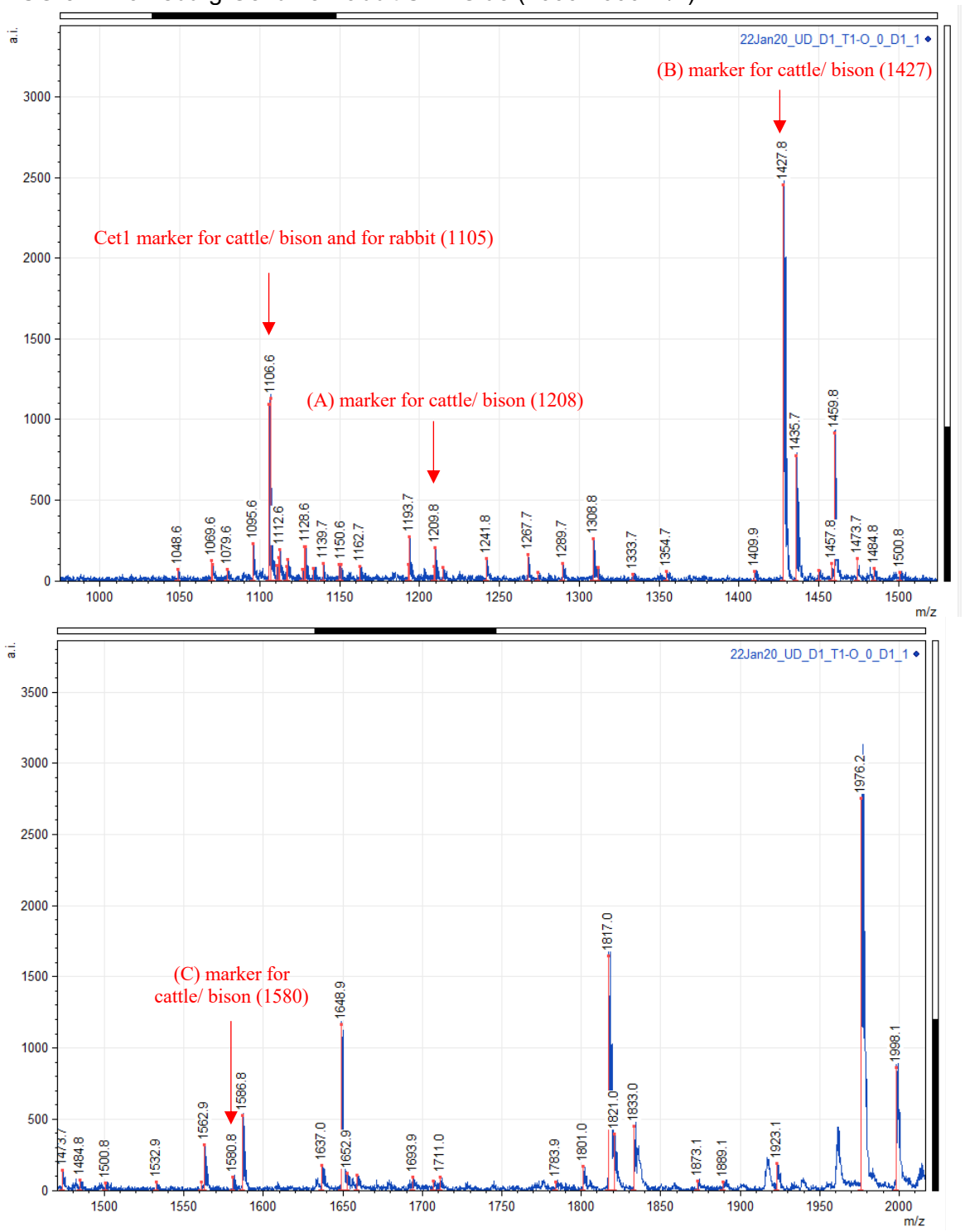


Figure 10. MALDI-TOF MS spectra for RSG2–*Kremer Pigments Rabbit Skin Glue Cubes* 1000-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison except for (G) which is not needed for identification. There is no indication of a mixture.

RSG-3 Williamsburg Genuine Rabbit Skin Glue (1000-2000 m/z)



RSG-3 Williamsburg Genuine Rabbit Skin Glue (2000-3200 m/z)

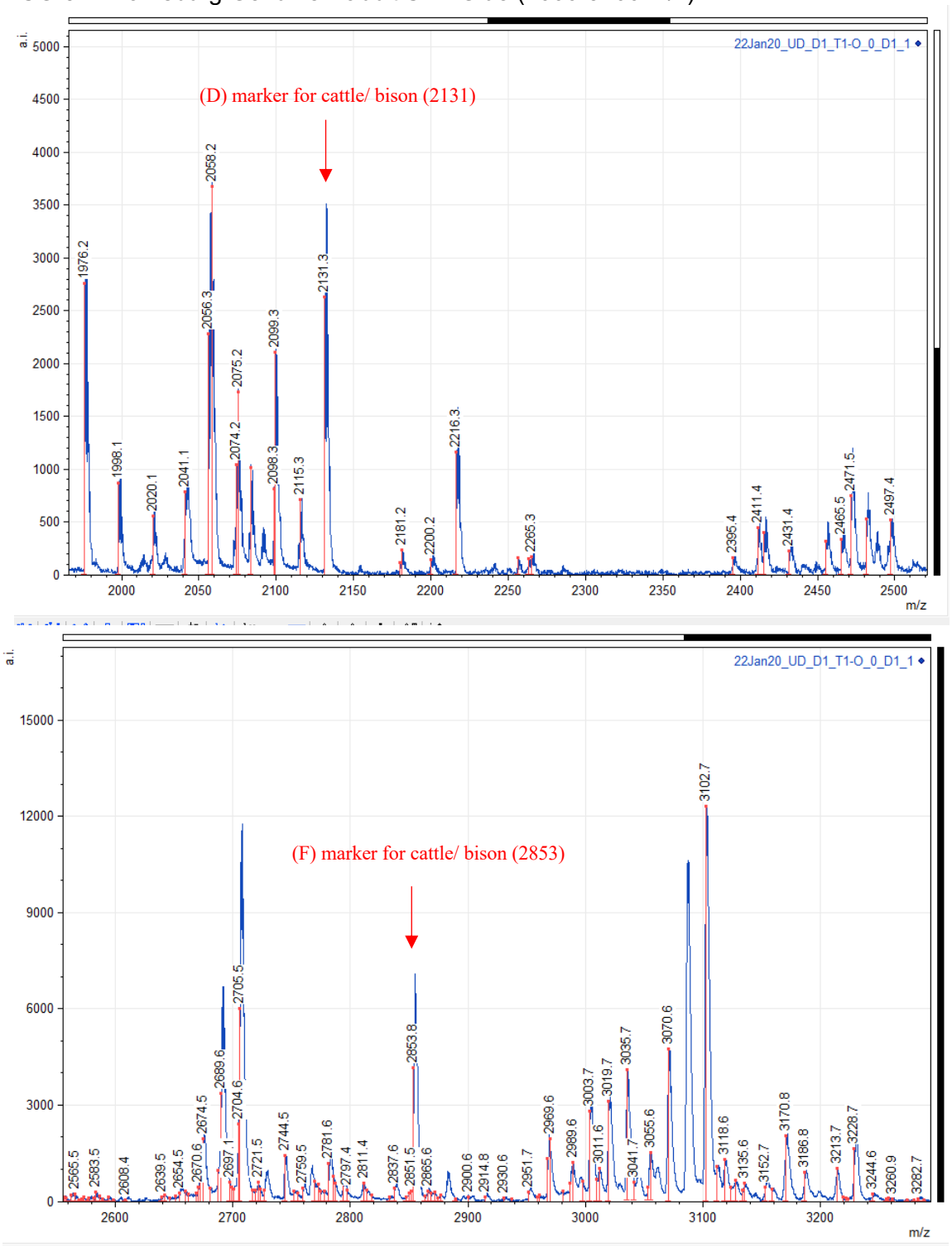
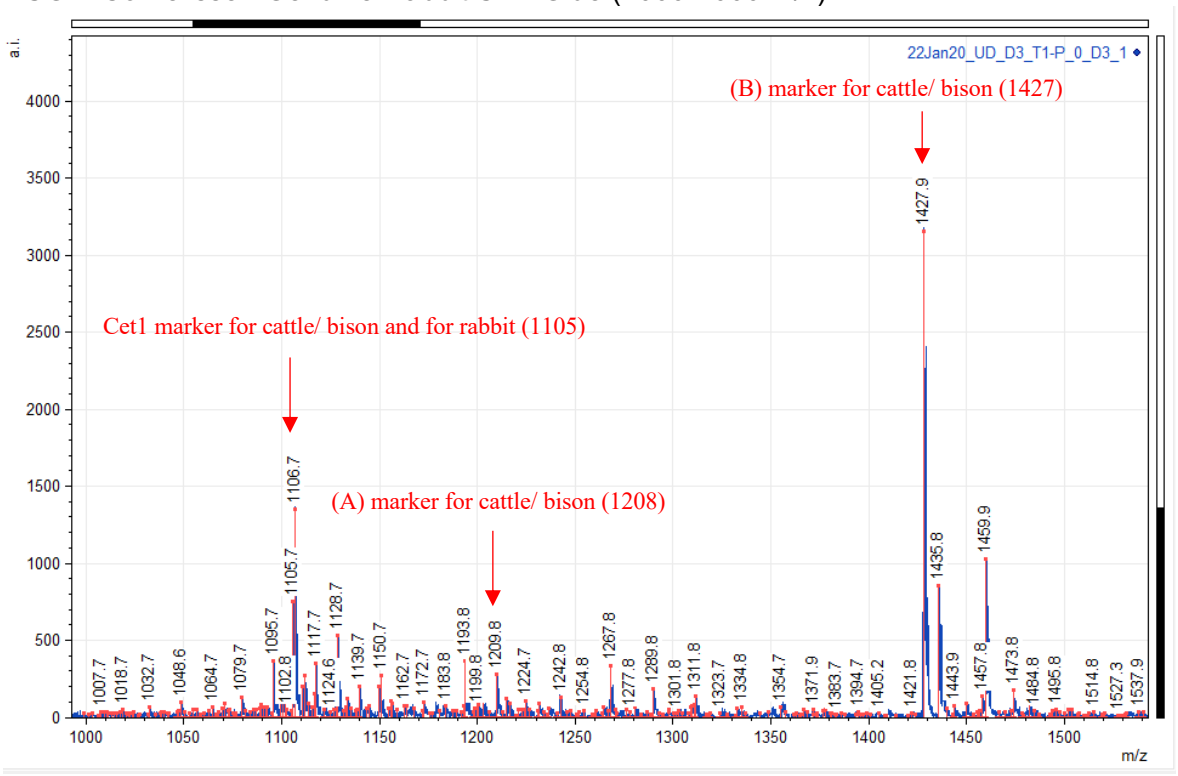
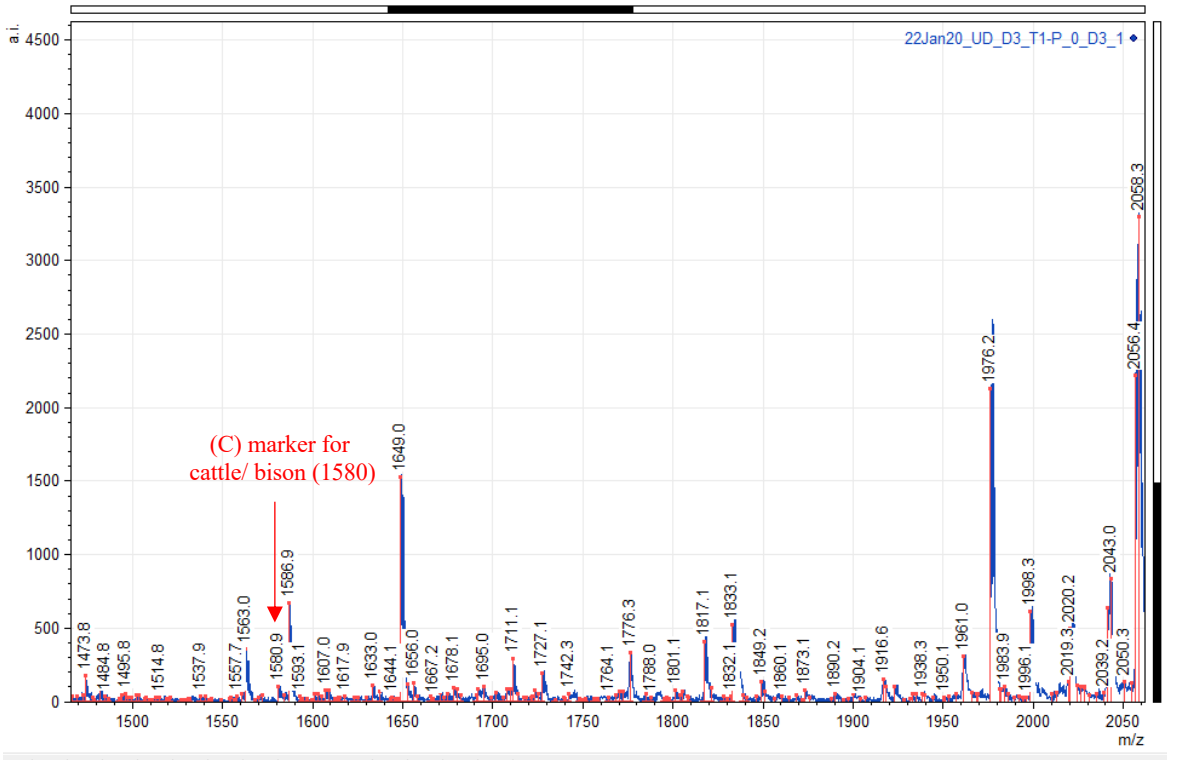


Figure 11. MALDI-TOF MS spectra for RSG3–*Williamsburg Genuine Rabbit Skin Glue* 1000-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison except for (G) which is not needed for identification. There is no indication of a mixture.

RSG-4 Cornelissen Genuine Rabbit Skin Glue (1000-2000 m/z)





RSG-4 Cornelissen Genuine Rabbit Skin Glue (2000-3200 m/z)

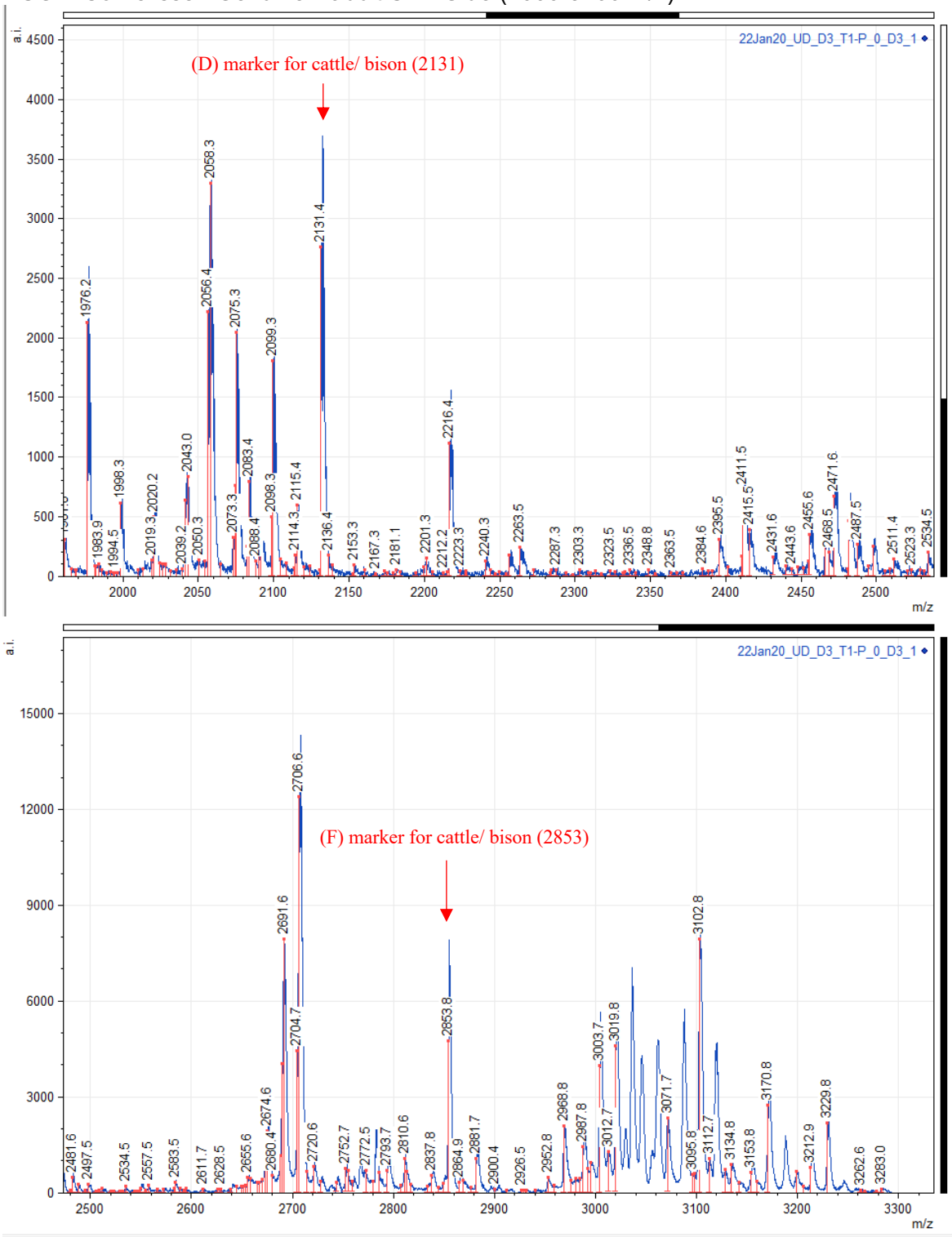
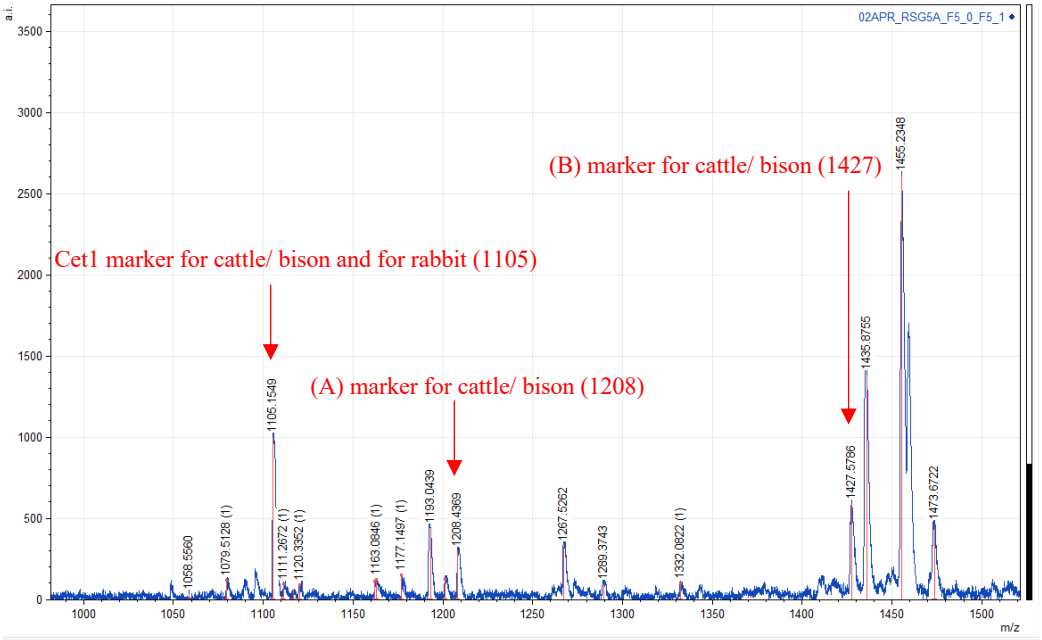
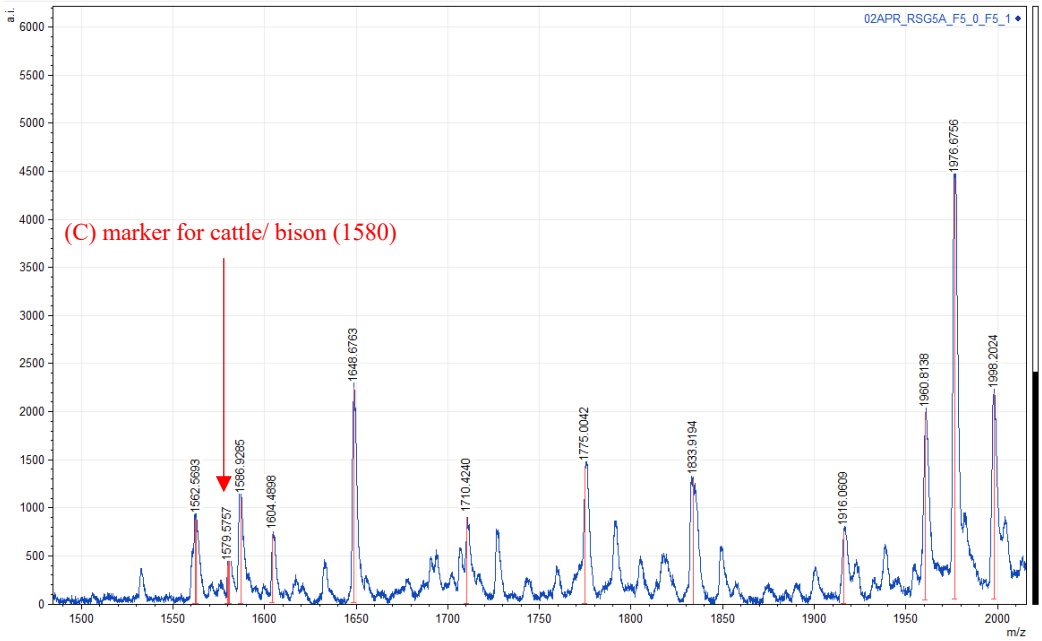


Figure 12. MALDI-TOF MS spectra for RSG–4 *Cornelissen Genuine Rabbit Skin Glue* 1000-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison except for (G) which is not needed for identification. There is no indication of a mixture.

RSG-5 Rublev/ Natural Pigments Genuine Rabbit Skin Glue (1000-2000 m/z)





RSG-5 Rublev/ Natural Pigments Genuine Rabbit Skin Glue (2000-3200 m/z)

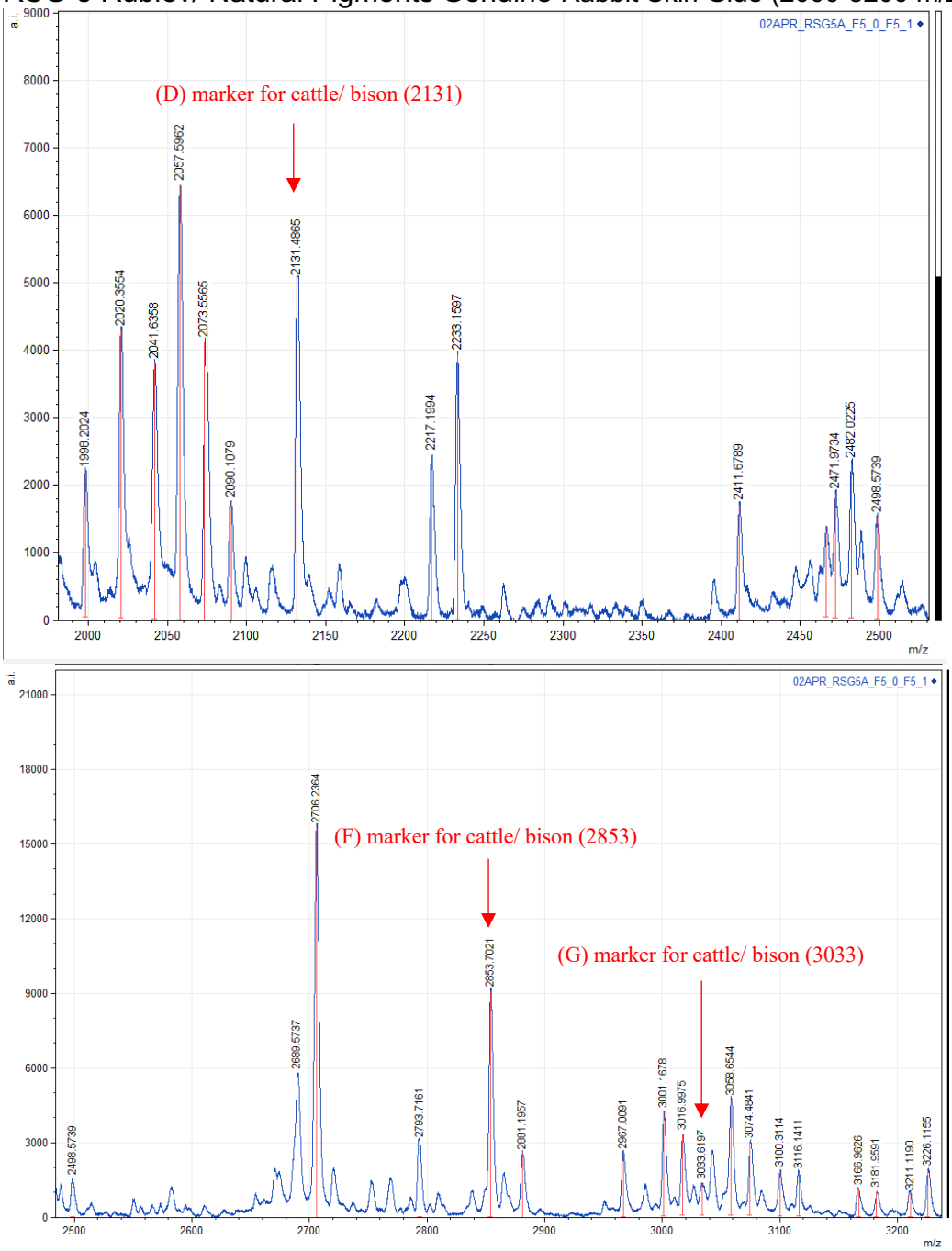
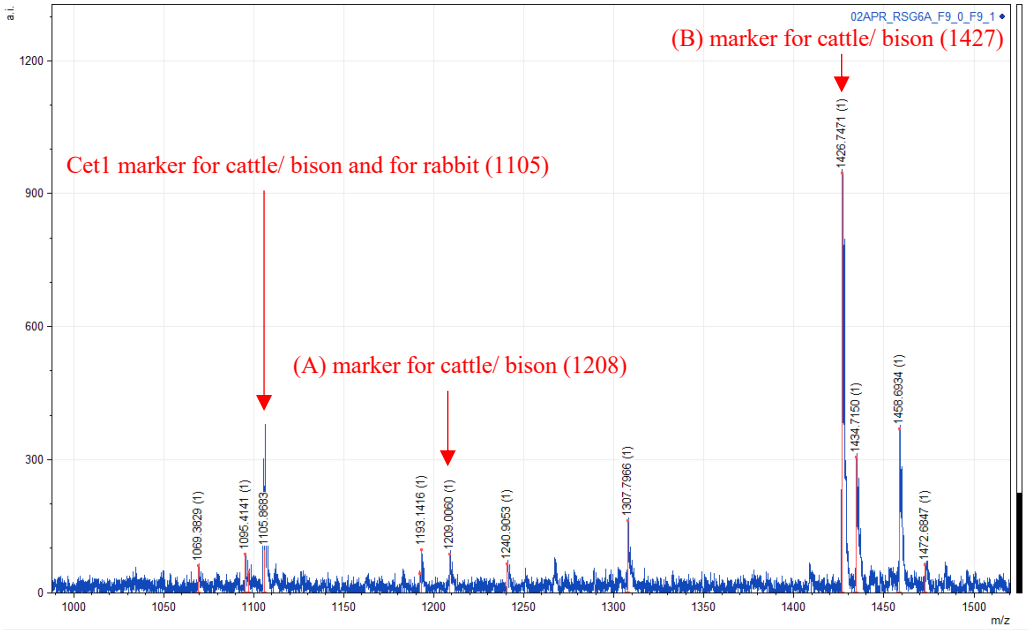
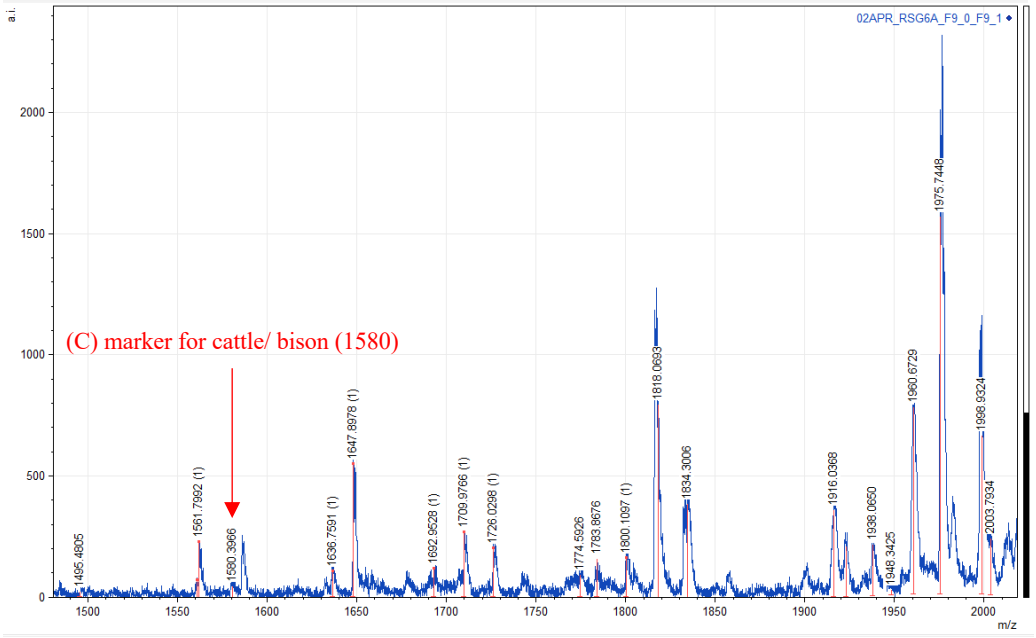


Figure 13. MALDI-TOF MS spectra for RSG5–*Rublev/ Natural Pigments Rabbit Skin Glue and Size* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison. There is no indication of a mixture.

RSG-6 Greg Dorrance Rabbit Skin Glue, German Granular (1000-2000 m/z)





RSG-6 Greg Dorrance Rabbit Skin Glue, German Granular (2000-3200 m/z)

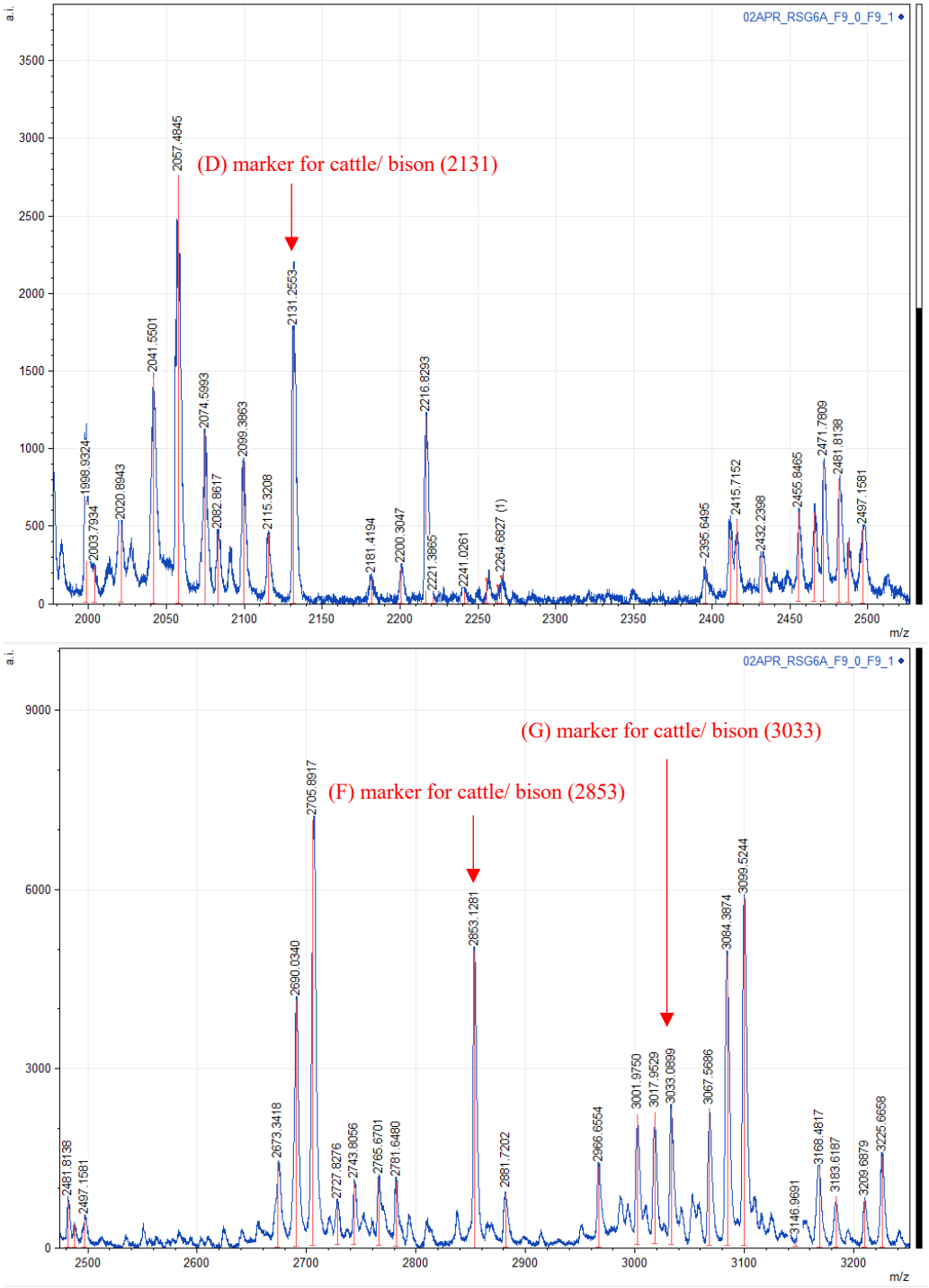
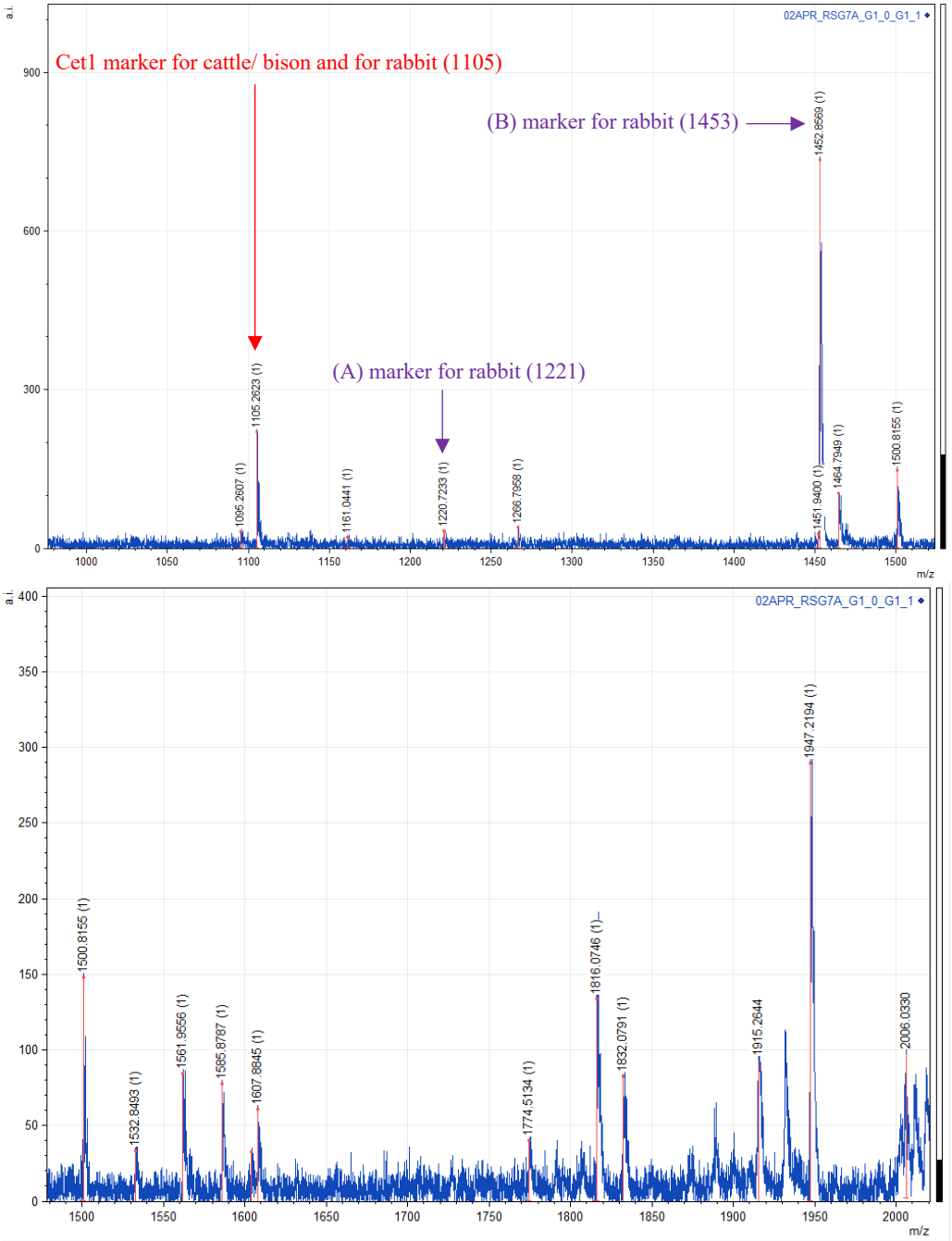


Figure 14. MALDI-TOF MS spectra for RSG6–*Greg Dorrance Rabbit Skin Glue, German Granular* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison. There is no indication of a mixture.

RSG-7 Woodfinishing Enterprises Imported Rabbit Skin Glue (1000-2000 m/z)



RSG-7 Woodfinishing Enterprises Imported Rabbit Skin Glue (2000-3200 m/z)

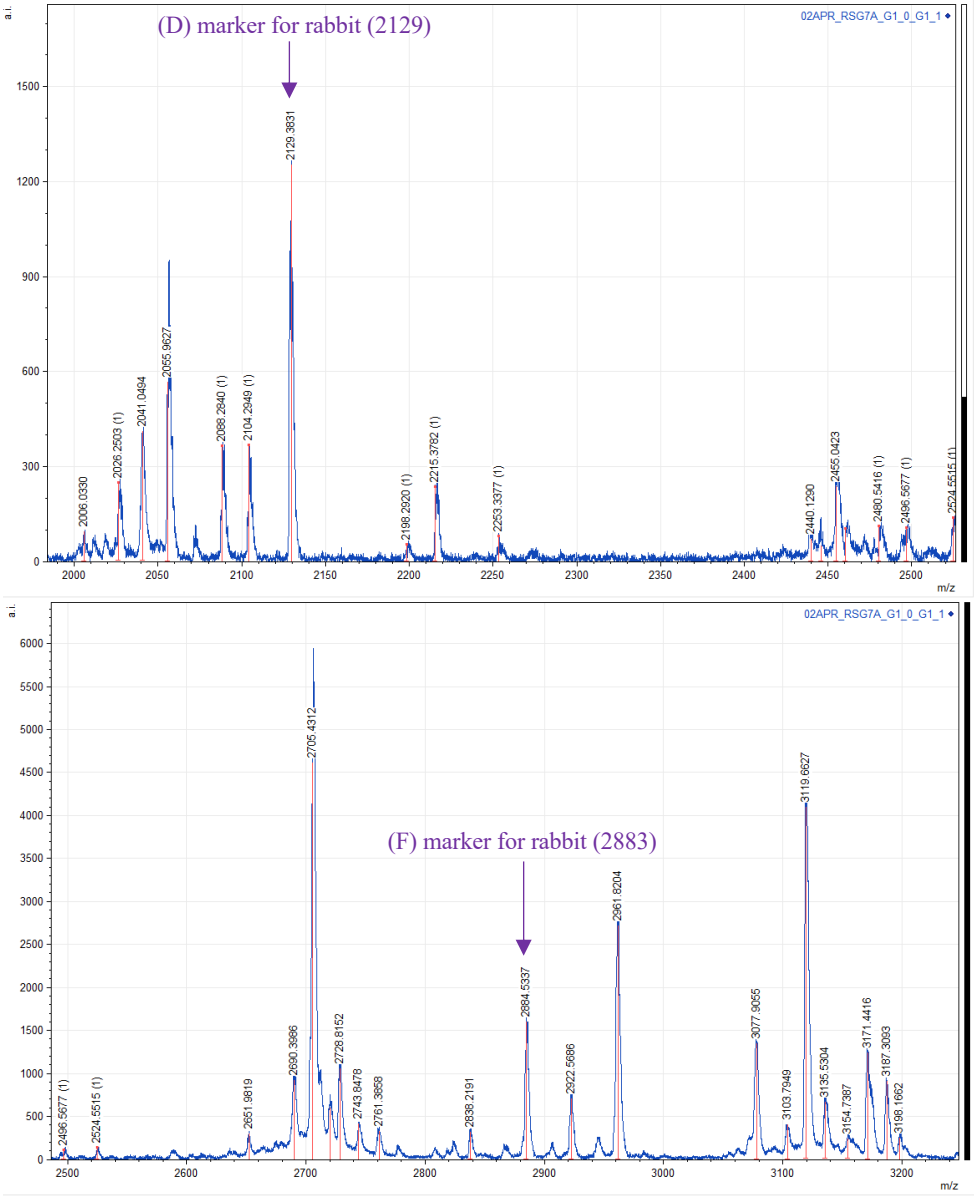
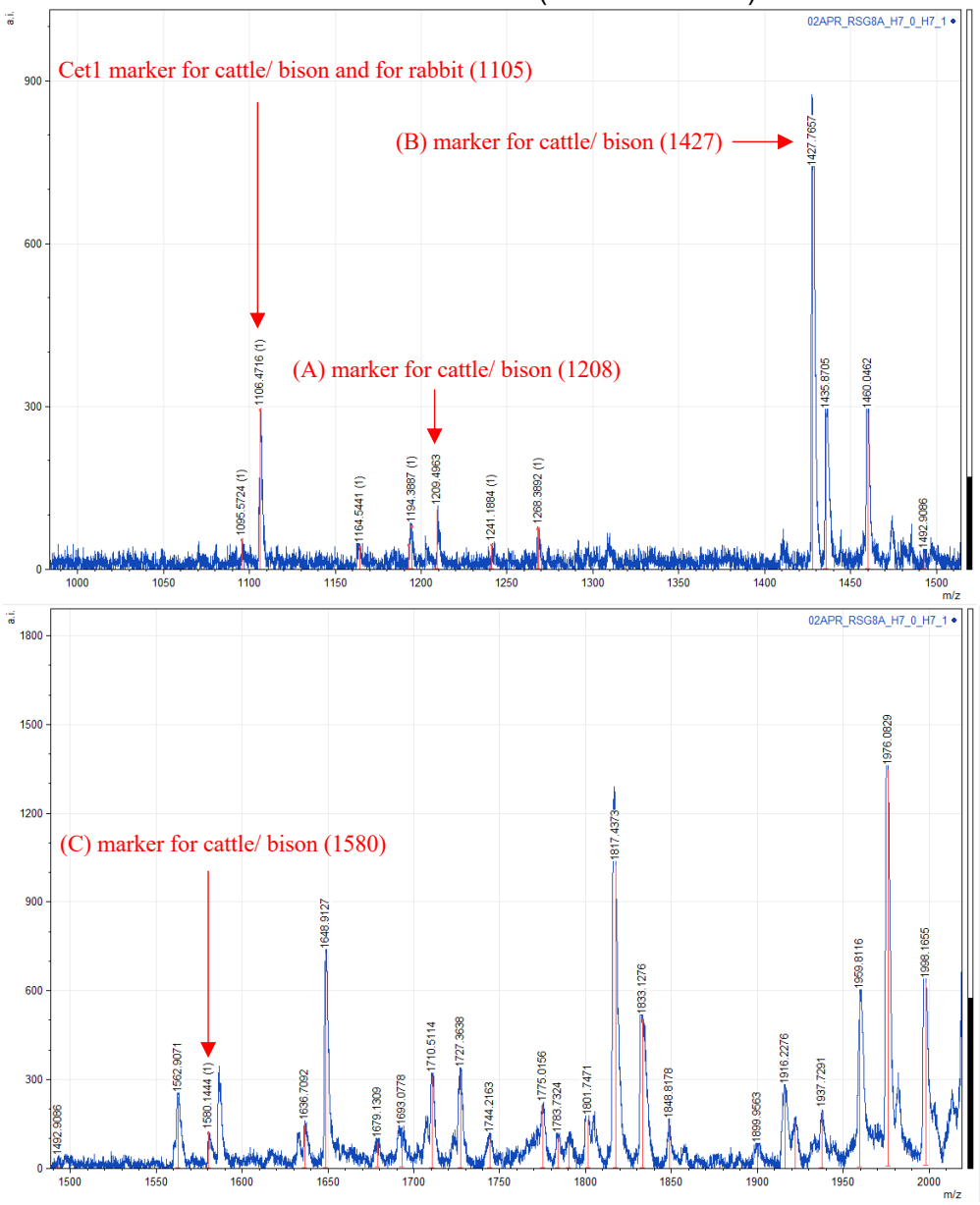


Figure 15. MALDI-TOF MS spectra for RSG7–*Woodfinishing Enterprises Imported Rabbit Skin Glue,* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with rabbit except for (C) and (G) which are not needed for identification. There is no indication of a mixture.

RSG-8 A.S. Handover Rabbit Skin Glue (1000-2000 m/z)



RSG-8 A.S. Handover Rabbit Skin Glue (2000-3200 m/z)

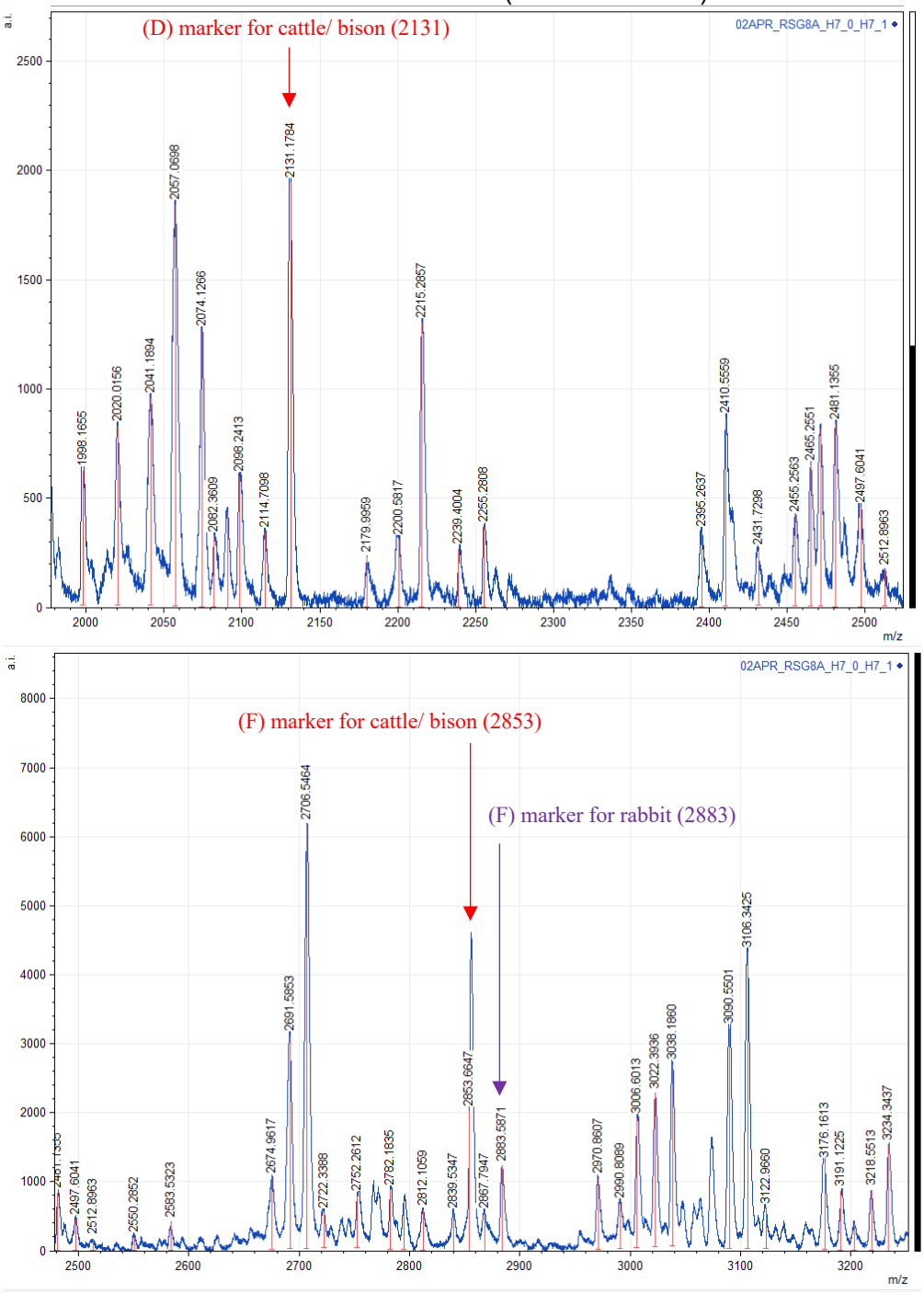
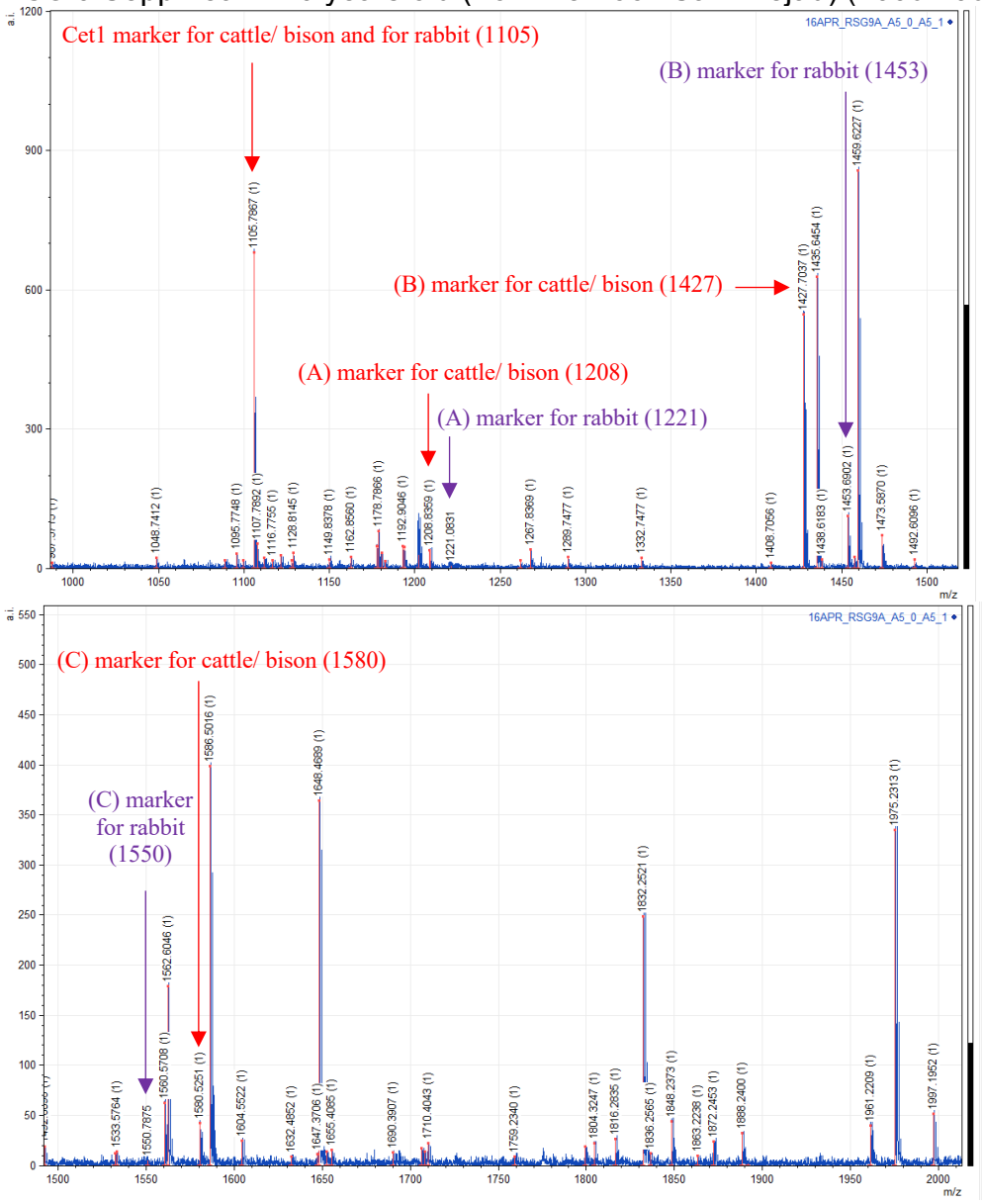


Figure 16. MALDI-TOF MS spectra for RSG8–*A.S. Handover Rabbit Skin Glue,* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison except for (G), which is not necessary for identification. There are two (F) markers, indicating a mixture. (F) marker 2883 is shared by many species, including rabbit. No other rabbit markers present.

RSG-9 Sepp Leaf: ~10 years old (from Behrooz Salimnejad) (1000-2000 m/z)



RSG-9 Sepp Leaf: ~10 years old (from Behrooz Salimnejad) (2000-3200 m/z)

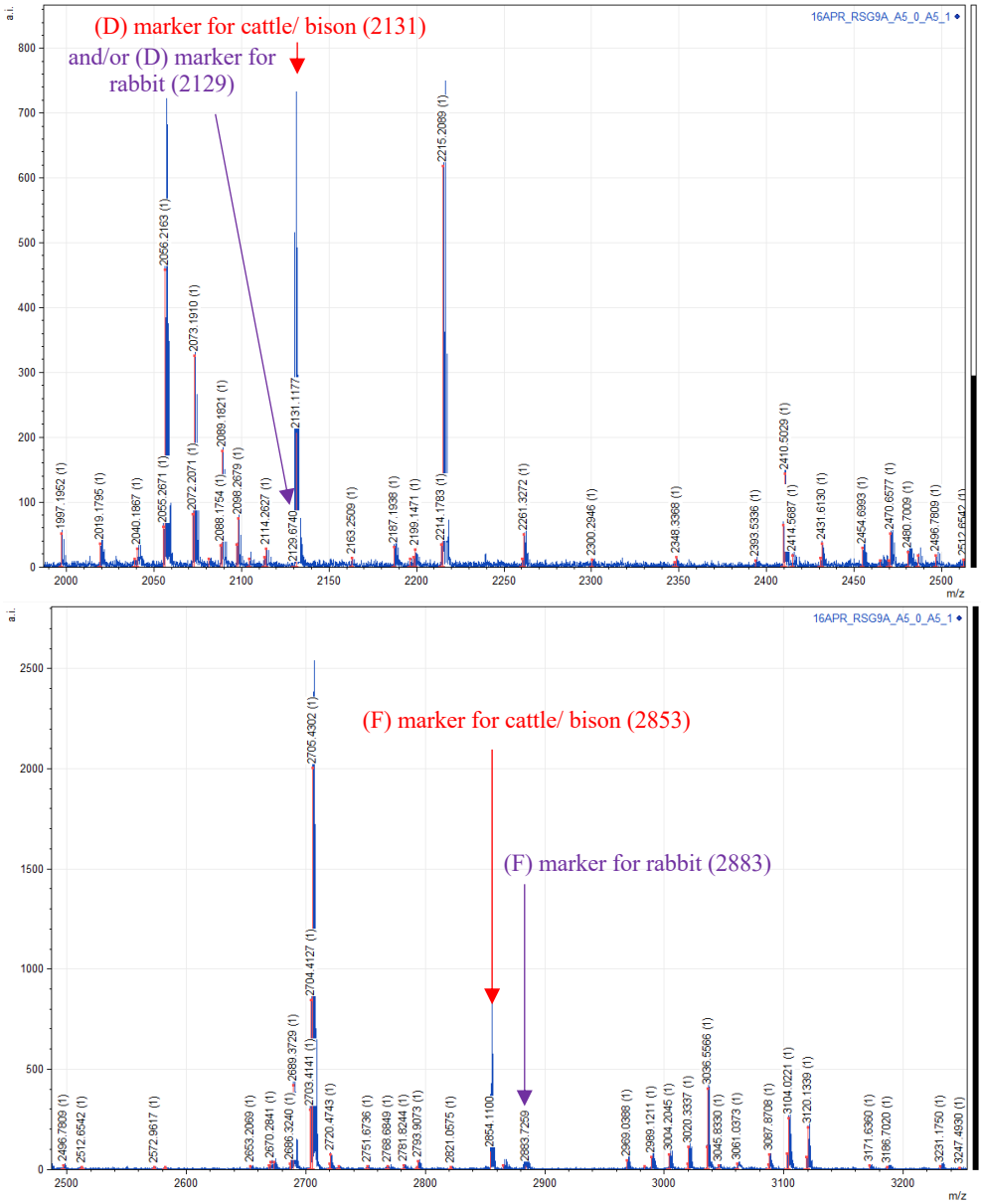
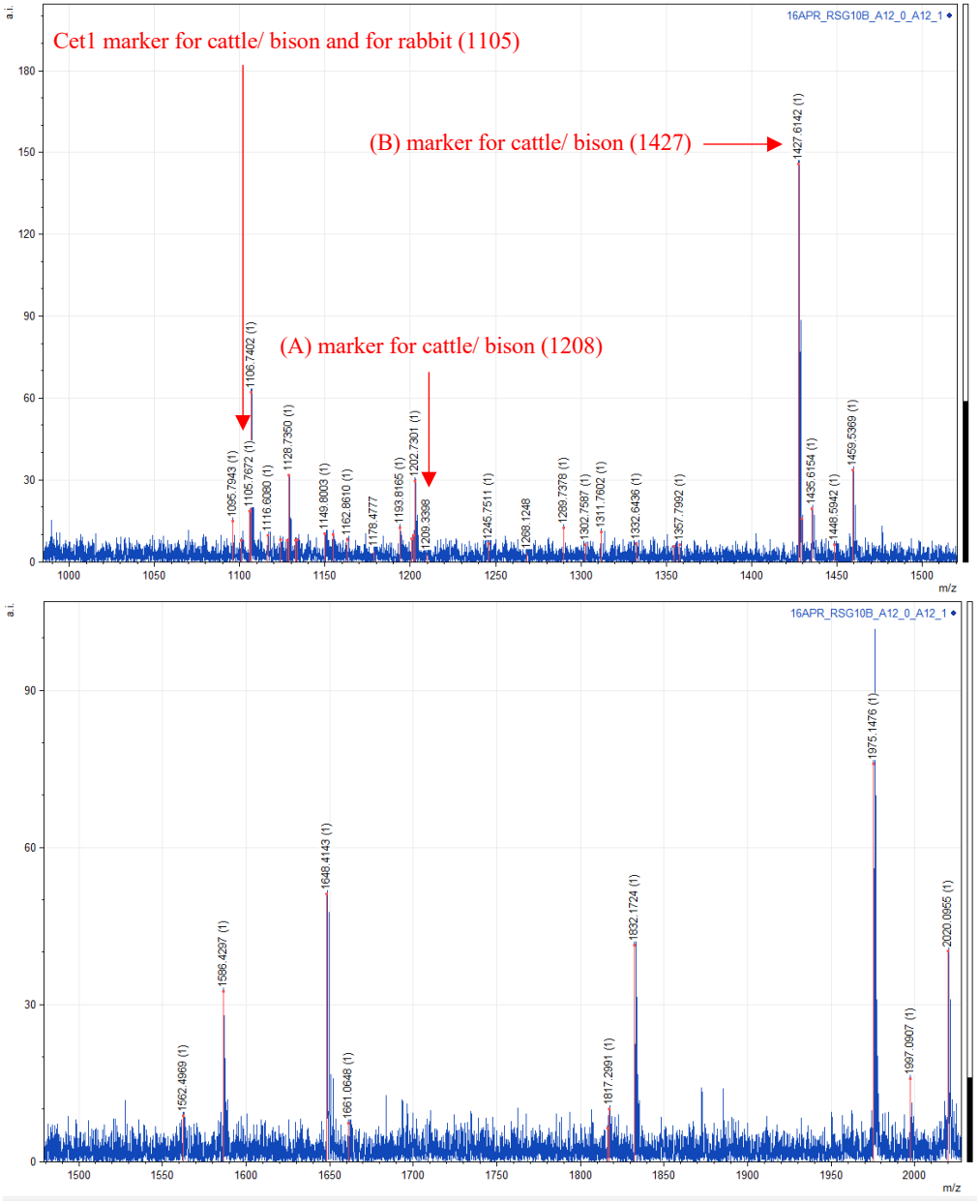


Figure 17. MALDI-TOF MS spectra for RSG9– *Sepp Leaf: ~10 years old (from Behrooz Salimnejad),* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison except for (G), which is not needed for identification. Shares all markers with rabbit except for (G), which is not needed for identification. Presence of two markers for all markers except for (G) indicates a mixture of rabbit and cattle/ bison. (D) markers for rabbit and for cattle/ bison are very close to each other and are difficult to resolve with certainty.

RSG-10 Sepp Leaf: ~3 years old (from Behrooz Salimnejad) (1000-2000 m/z)



RSG-10 Sepp Leaf: ~3 years old (from Behrooz Salimnejad) (2000-3200 m/z)

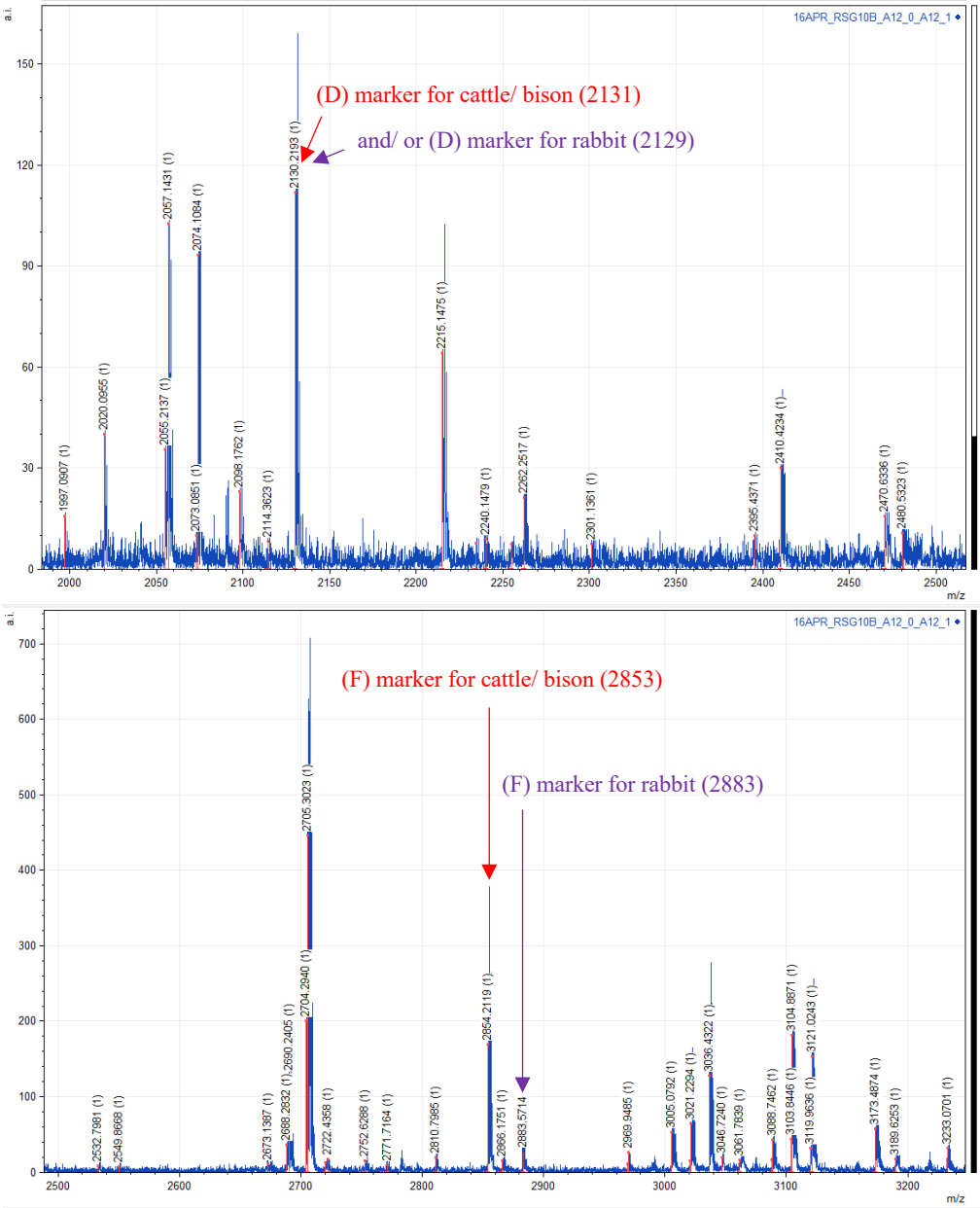
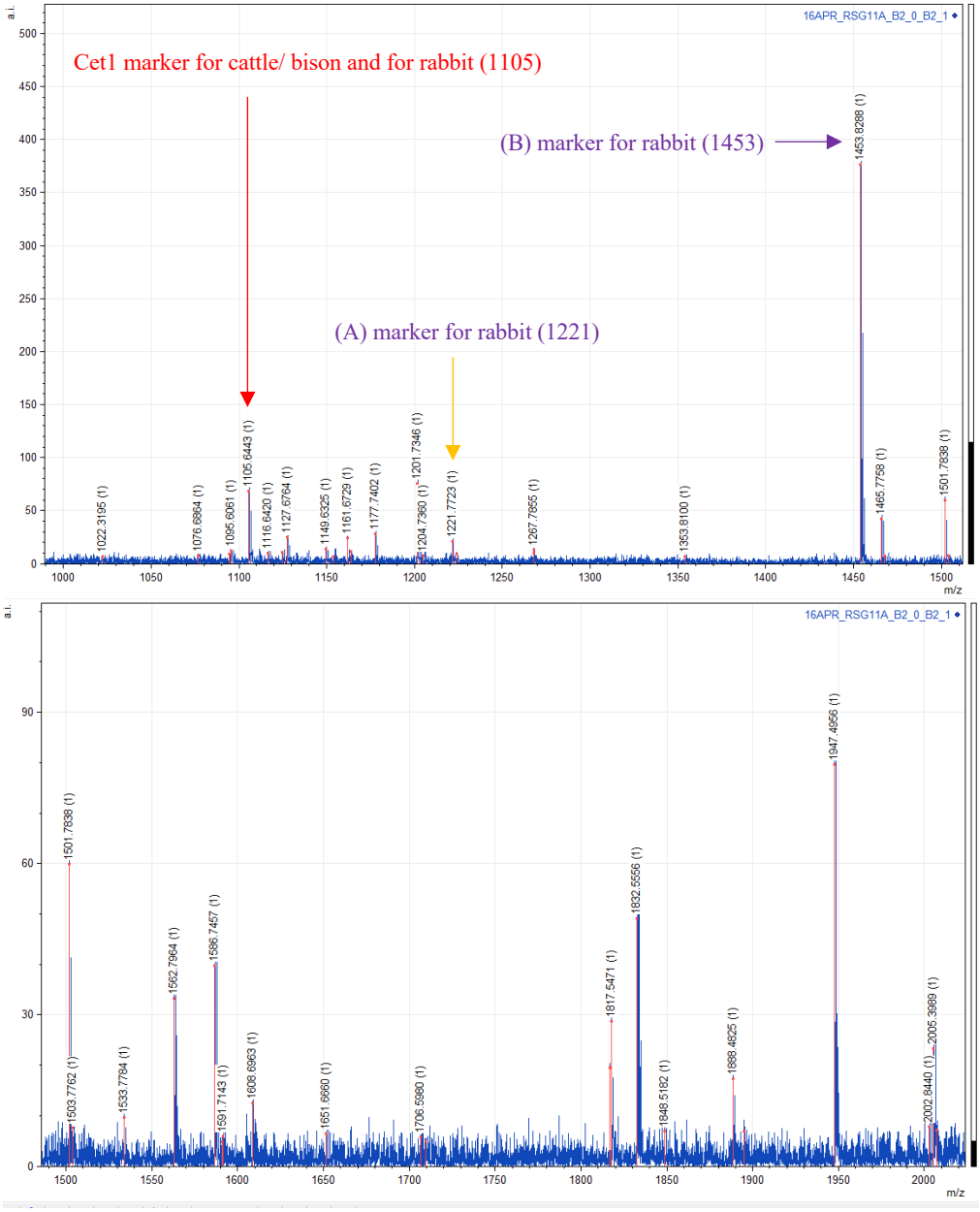


Figure 18. MALDI-TOF MS spectra for RSG10– *Sepp Leaf: ~3 years old (from Behrooz Salimnejad),* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison except for (C) and (G), which are not needed for identification. There are two (F) markers, indicating a mixture. (F) marker 2883 is shared by many species, including rabbit. Could have (D) marker for rabbit; (D) markers for rabbit and for cattle/ bison are very close to each other and are difficult to resolve with certainty.

RSG-11 Iranian Rabbit Skin Glue (from Behrooz Salimnejad) (1000-2000 m/z)



RSG-11 Iranian Rabbit Skin Glue (from Behrooz Salimnejad) (2000-3200 m/z)

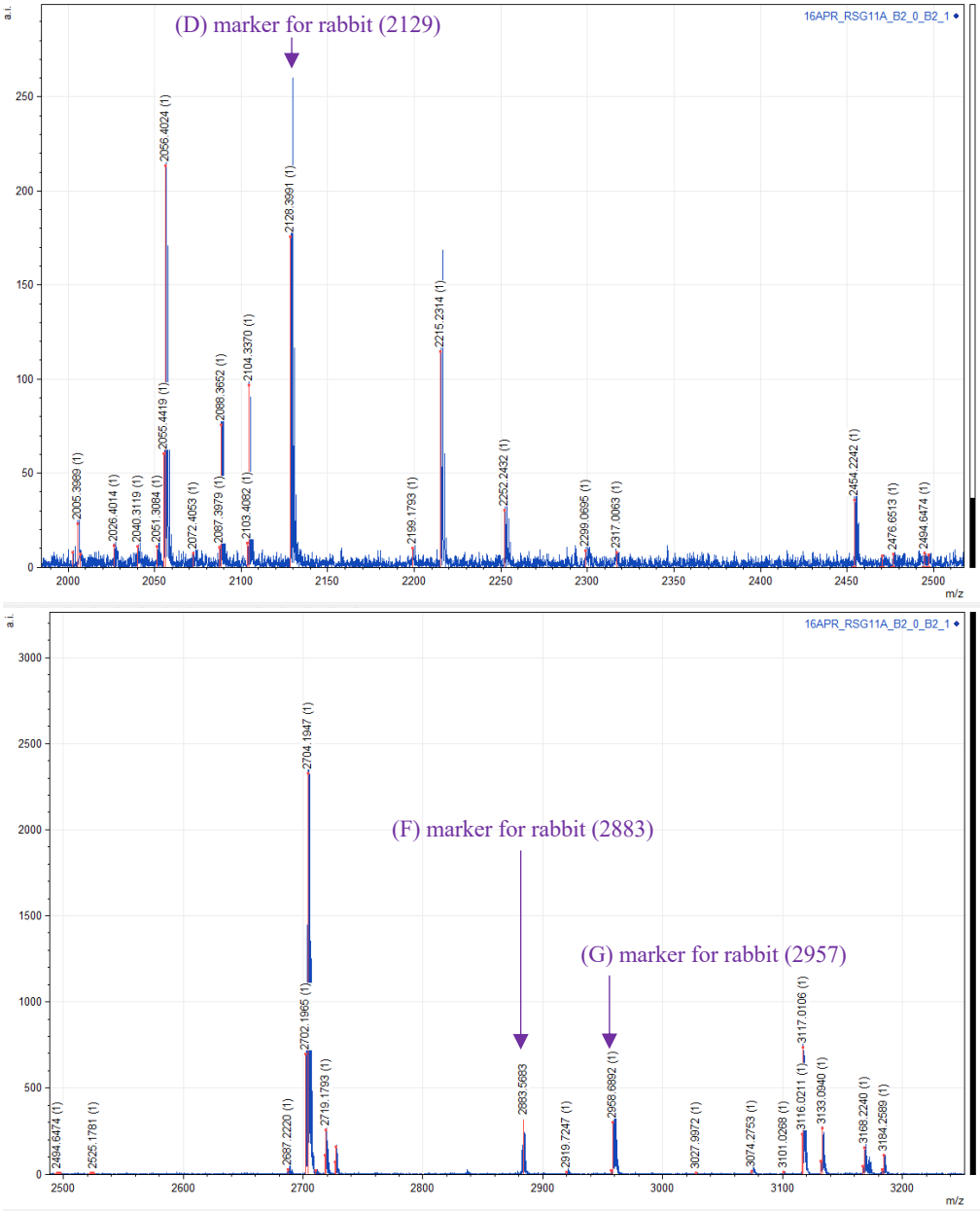
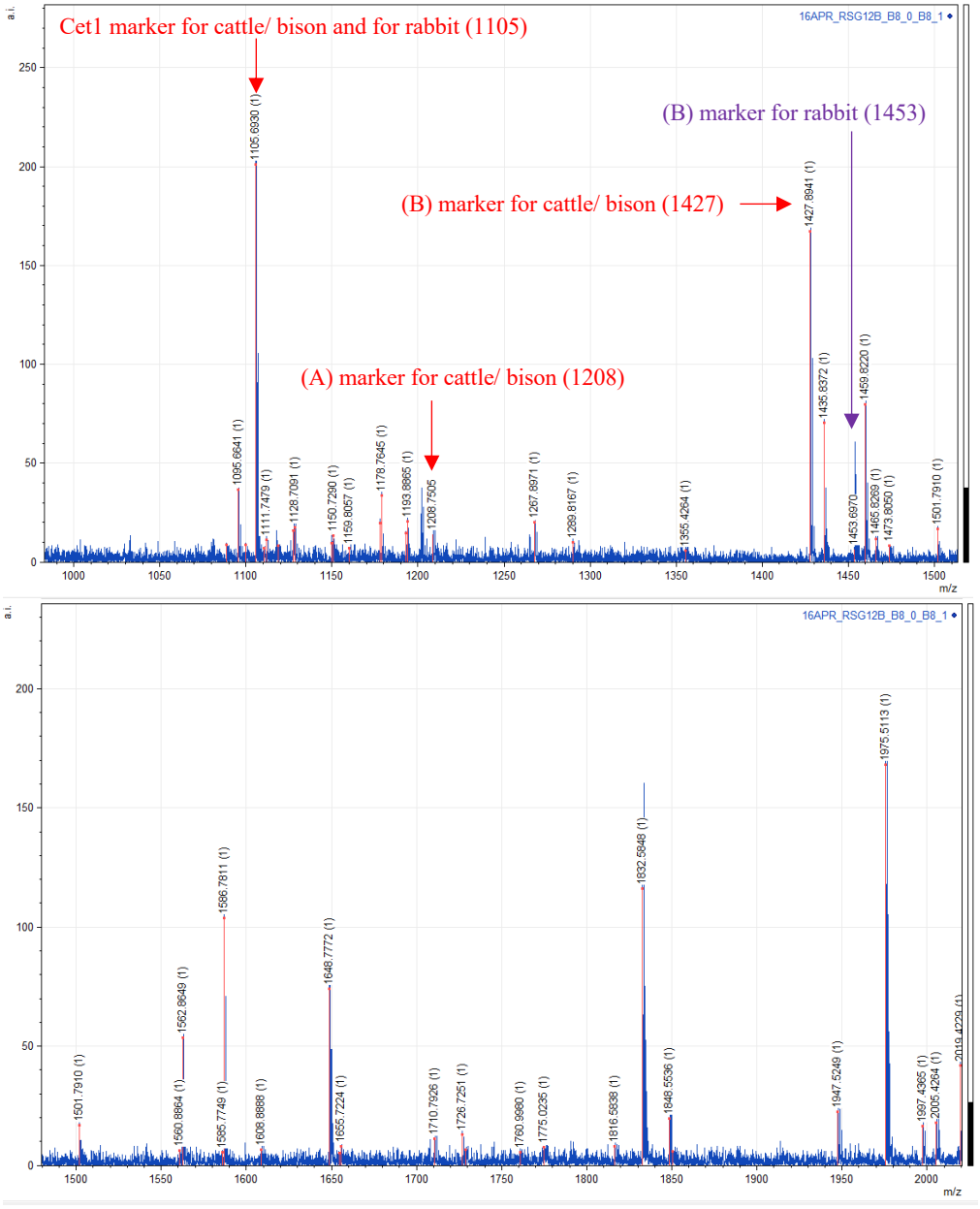


Figure 19. MALDI-TOF MS spectra for RSG11– *Iranian Rabbit Skin Glue (from Behrooz Salimnejad),* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with rabbit except for (C) which is not needed for identification. There is no indication of a mixture.

RSG-12 German Rabbit Skin Glue (from Behrooz Salimnejad) (1000-3200 m/z)



RSG-12 German Rabbit Skin Glue (from Behrooz Salimnejad) (2000-3200 m/z)

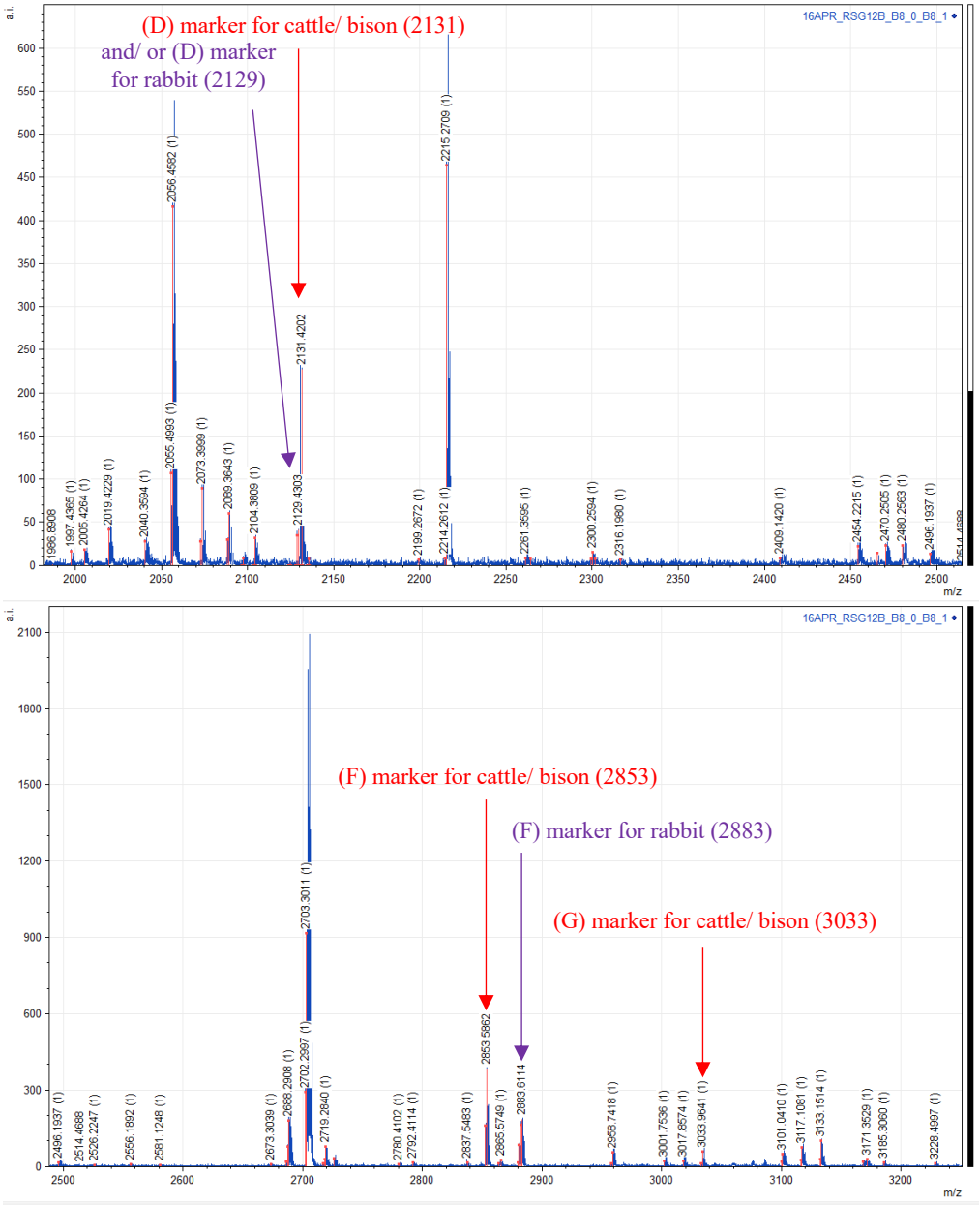
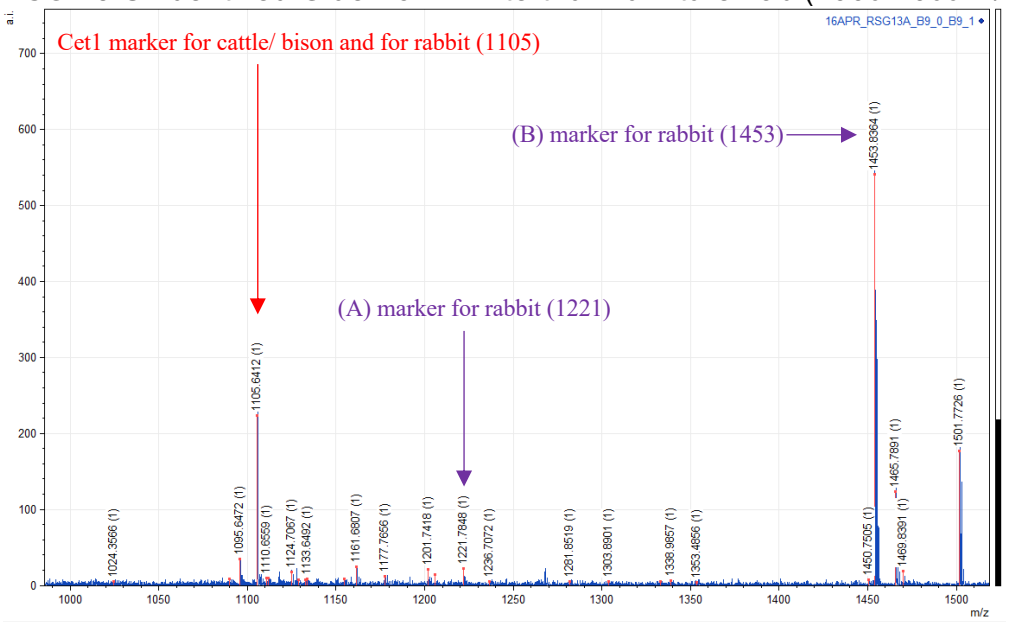
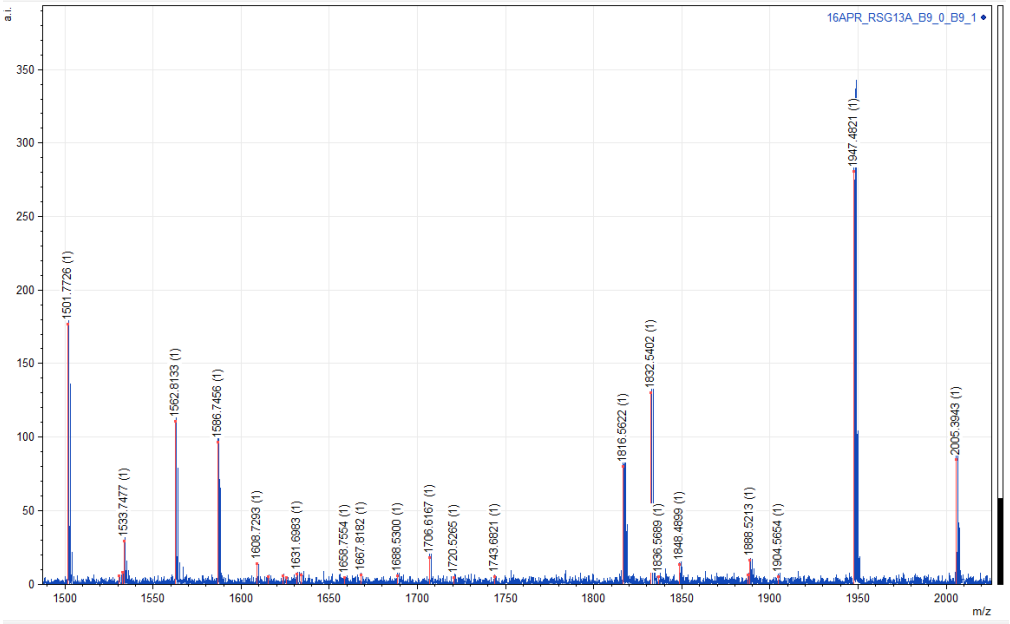


Figure 20. MALDI-TOF MS spectra for RSG12– *German Rabbit Skin Glue (from Behrooz Salimnejad),* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with cattle/ bison except for (C), which is not necessary for identification. Shares (B), (F), and possibly (D) markers with rabbit. (D) markers for rabbit and for cattle/ bison are very close to each other and are difficult to resolve with certainty.

RSG-13 Unidentified Glue from Winterthur Furniture Lab (1000-2000 m/z)





RSG-13 Unidentified Glue from Winterthur Furniture Lab (2000-3200 m/z)

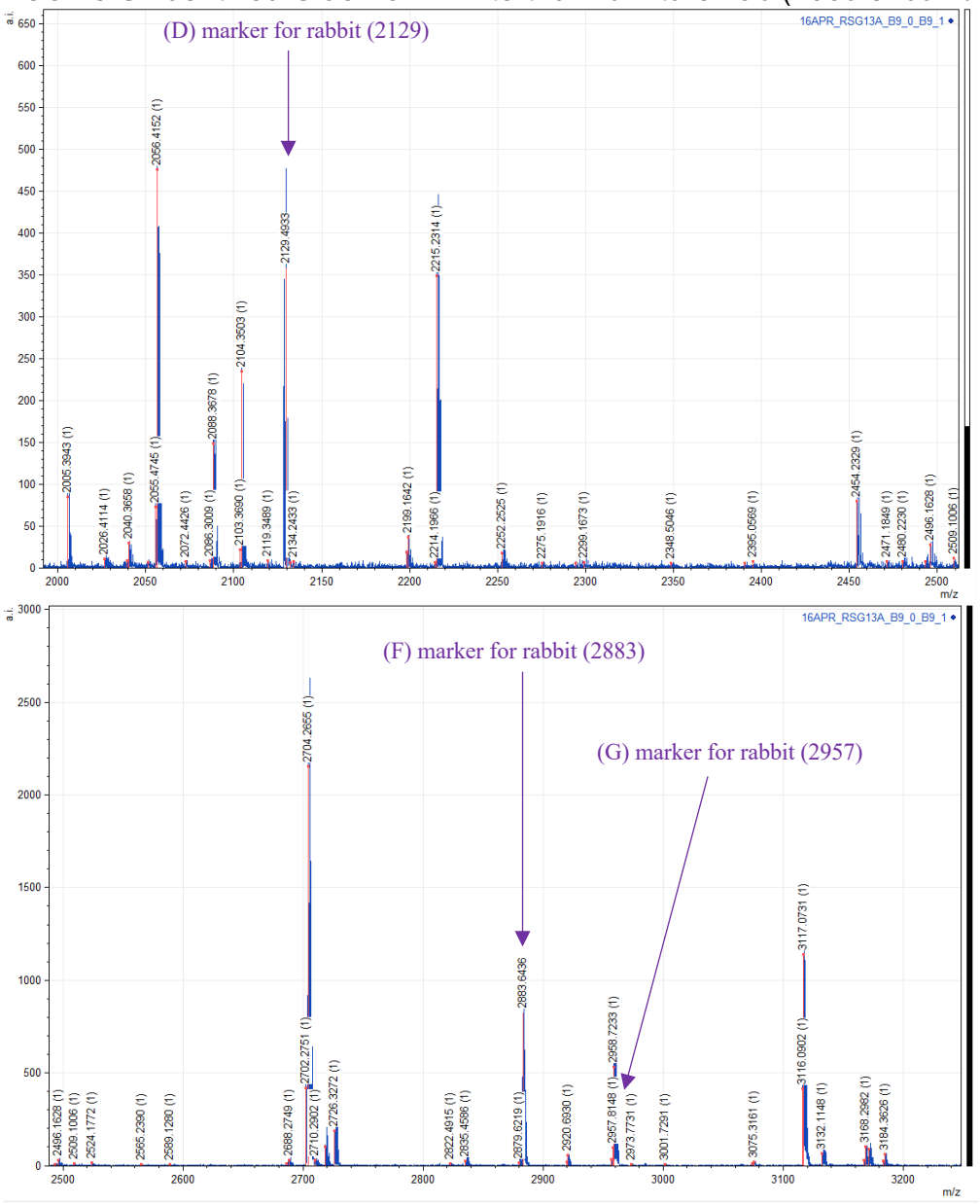


Figure 21. MALDI-TOF MS spectra for RSG13– *Unidentified Glue from Winterthur Furniture Lab,* 0-3200 m/z (this page and previous page–spectra divided into four sections for legibility). Shares all markers with rabbit except for (C) which is not needed for identification. There is no indication of a mixture.

Updated 7/10/2017	Cet1	(A)	(B)	(C)	Cet2	(D)	(F)	(G)	Other
Walrus	1105	1221	1453	1566	1652	2121	2853	3003	
Northern fur seal	1105	1221	1453	1566	1652	2121	2853	2957	
Stellar sea lion	1105	1221	1453	1566		2121	2853	2957	
Bearded seal	1121	1221	1453	1566	1652	2171	2853	2957	
Ringed seal*	1105	1221	1453	1566	1652	2171	2869	2957	
Phocini seal (ribbon, spotted, grey, harbor and harp seal)	1105	1221	1453	1566	1652	2171	2869	2957	
Hooded seal	1105	1221	1453	1566	1652	2171	2853	2957	
Cattle / Bison	1105	1208	1427	1580		2131	2853	3033	
Sheep/Pronghorn	1105	1196	1427	1580		2131	2883	3033	
Goat	1105	1196	1427	1580		2131	2883	3093	
Musk Ox	1105	1208	1427	1580		2131	2883	3033	
Elk/Red Deer/Fallow deer	1105	1196	1427	1550		2131	2883	3033	
Caribou/Reindeer	1105	1166	1427	1580		2131	2883	3093	
Roe Deer	1105	1196	1427	1550		2131	2883	3059	
N. A Deer: Mule, Sitka, Whitetail	1105	1196	1427	1580		2131	2883	3059	
Horse	1105	1198	1427	1550	1682	2145	2883	2999	
Common / Bottlenose / White-beaked / Euphrosyne dolphin	1079	1205	1453	1566	1638	2119	2883	3023	
Risso's Dolphin / Pilot whale / false killer whale	1063	1205	1453	1566	1638	2119	2883	3023	
Orca / White-sided dolphin	1079	1205	1453	1566	1652	2119	2883	3023	
Porpoise	1079	1205	1453	1550	1652	2119	2883	3023	
Narwhal	1079	1205	1443	1550	1652	2089	2883	3051	
Beluga whale	1079	1205	1443	1550	1652	2121	2883	3051	
Sperm whale	1079	1205	1453	1550	1652	2133	2883	3039	
Bottlenose / Sowerby's whale	1063	1205	1441	1550	1638	2091	2883	3023	
Minke whale	1079	1205	1441	1566	1652	2135	2883	3023	
Fin Whale	1079	1205	1453	1566	1652	2135	2883	3023	
Humpback whale	1079	1205	1453	1566	1652	2135	2869	3023	
Blue whale	1079	1205	1453	1550	1652	2105	2883	3023	
Gray whale	1079	1205	1453	1566	1652	2135	2899	3023	
Sei whale	1079	1205	1441	1550	1652	2135	2883	3023	
Right whale	1079	1205	1453	1566	1682	2135	2883	3023	
Elephant	1105	1251	1453	1579	1540	2115	2853	2999/3015	3048?
Mastodon	1105	1251	1453	1579	1568	2115	2853	2999/3015	3048?
Black rhino	1105	1184/1198	1453	1550		2145	2869	2999	
Red fox	1105	1210/1226	1437	1566		2131	2853	2999	
Arctic fox	1105	1210/1226	1453	1566		2131	2853	2999	
Gray fox	1105	1208/1224	1453	1566		2131	2853	2899	
Cat	1105	1207/1223	1453	1566		2163	2853	2999	
Pig	1105/1121	1180/1196	1453	1550		2131	2883	3033	
Rabbit	1105	1221/1235	1453	1550?		2129	2883	2957	
Rat	1105	1187/1203	1453	1566		2143	2883	3003	
Mouse	1105	1178/1194	1453	1566		2159	2883	2947	
Water bullalo	1105	1192/1208	1455	1580		2131	2883/2899	3059/3075	
Dog/wolf	1105	1226	1453	1566		2131	2853	2999	
Bear: Brown/Black/Polar	1105	1233	1453	1566		2163	2853	2957	
Lion	1105	1223	1453	1566		2147	2853	2999	
Lynx	1105	1223	1453	1566		2147	2853	2957	
Human	1105	1235	1477	1580		2115	2869	2957	2832?

Figure 22. PMF markers used to identify collagen-based materials (courtesy of Dr. Dan Kirby).