ACS SENSORS
Article
pubs.acs.org/acssensors

Differentiation and Identification of Cachaça Wood Extracts Using Peptide-Based Receptors and Multivariate Data Analysis

Eman Ghanem,[†,#] Saazina Afsah,[‡,¶] Parisa N. Fallah,[‡,¶] Alexandria Lawrence,[‡,¶] Elise LeBovidge,[‡,¶] Sneha Raghunathan,[‡,¶] Dominic Rago,[‡,¶] Michelle A. Ramirez,[‡,¶] Mitchell Telles,[‡,¶] Michelle Winkler,[‡,¶] Bailey Schumm,[‡] Kiran Makhnejia,[‡] David Portillo,[‡] Rafael C. Vidal,[‡] Allison Hall,[‡] Debbie Yeh,[‡] Holly Judkins,[‡] Alexandre Ataide da Silva,[§] Douglas W. Franco,[§,△] and Eric V. Anslyn[*,∥]

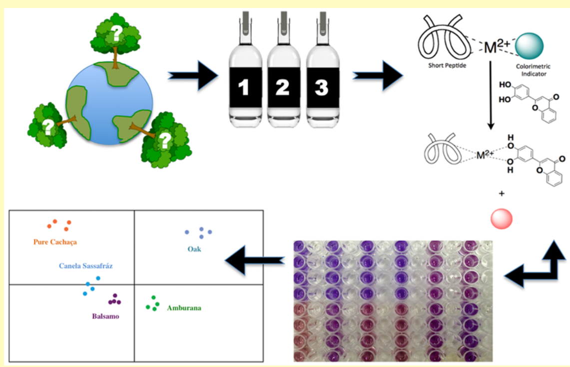
[†]Texas Institute for Discovery Education in Science, College of Natural Sciences and [‡]Freshman Research Initiative, The University of Texas at Austin, Mail Stop G2550, Austin, Texas 78712, United States

[§]Departamento de Química e Física Molecular, Instituto de Química de São Carlos, Universidade de São Paulo (USP), Av. Trabalhador Sancarlense 400, CP 780, CEP 13560-970, São Carlos, SP, Brazil

[∥]Department of Chemistry, The University of Texas at Austin, 105 E 24th Street, Mail Stop A5300, Austin, Texas 78712-1224, United States

Supporting Information

ABSTRACT: It is becoming increasingly important to differentiate complex mixtures, especially in forensics. Cachaça, the most popular alcoholic beverage in Brazil, is made from distilled and fermented sugar cane juice. It contains a mixture of naturally occurring polyphenols known as tannins, whose composition is dictated by the type of wood used to age the beverage. These tannins can be differentiated in an Indicator Displacement Assay (IDA) using peptide-based ternary sensing ensembles. This investigation demonstrates a technique for fingerprinting the identity of the woods used to age cachaças. Unknown cachaça samples were tested against a training set of Brazilian woods in addition to oaks from different countries. Results obtained from the analysis showed that 62.5% of the samples were correctly identified. Furthermore, four samples anonymously added to the pool of unknowns from the training set were identified with 100% accuracy, emphasizing both the promising results obtained from this method of differentiation and the importance of analyzing same-age samples.



KEYWORDS: *Cachaça, wood identification, indicator displacement assay, differential sensing, multivariate data analysis, oak, Brazilian wood*

Cachaça, the most popular alcoholic beverage in Brazil, has been gaining popularity worldwide.[1] It is often confused with rum, but unlike rum, which is produced from molasses, cachaça is produced from distilled sugar cane juice. After fermentation and distillation in copper or stainless steel distillers,[2] the beverage is aged in oak or native Brazilian wooden casks to modify and enhance its sensory attributes.[3] Because oak is not a native tree to Brazil, producers import oak casks from Europe and North America. Recently, Brazilian woods such as amburana, amendoim, balsamo, and canela-sassafrás have been considered as alternatives to oak in aging cachaça.[4−7]

With the introduction of different Brazilian woods to cachaça production, there is an increasing demand for characterization and wood identification techniques to prevent adulteration and illegal logging of endangered species.[8] Traditional wood identification techniques such as scanning electron microscopy (SEM),[9] near-infrared (NIR) spectroscopy,[10] and ion-mobility spectrometry[11] require specialized equipment and facilities. More recently, an electronic nose and laser-induced breakdown spectrometry (LIBS) have been used to differentiate four Brazilian wood species based on their composition of volatile organic compounds and chemical elements, respectively.[12]

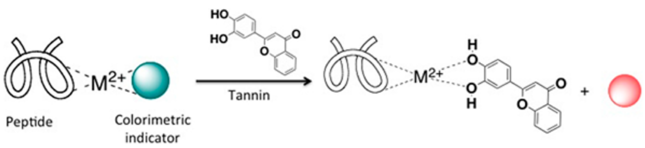
Differential sensing using arrays of cross-reactive receptors coupled with chemometric analysis is an emerging tool for the analysis of complex mixtures that does not require the identification of individual components in the mixture.[13] We previously developed a class of multicomponent, cross-reactive supramolecular sensors, and also demonstrated their applications in fingerprinting red wine varietals based on their composition of the naturally occurring polyphenols, tannins.[14] Our peptide-based sensing ensembles are composed of short histidine-containing peptides, a divalent metal ion, and a pH colorimetric indicator that contains structural motifs similar to

Received: December 12, 2016
Accepted: April 28, 2017
Published: April 28, 2017

DOI: 10.1021/acssensors.6b00809
ACS Sens. XXXX, XXX, XXX−XXX

tannins. Responses from the sensing ensembles to tannin binding are monitored using an analytical method known as indicator displacement assay (IDA).[15] IDA is based on the displacement of a colorimetric indicator by the analyte, causing a color change that can be measured spectrophotometrically (Scheme 1). In addition to their application in fingerprinting

Scheme 1. Schematic Representation of the Indicator Displacement Assay (IDA)



red wine varietals, we have demonstrated the utility of peptidic sensors in discriminating Cabernet Sauvignon wines based on grape maturation time[16] and in predicting the composition of red wine blends prepared by mixing three wine varietals.[17]

Inspired by our previous findings, herein we apply the peptide-based sensing array to the differentiation of cachaça extracts of oak and Brazilian woods. We tested five types of oak obtained from different countries in addition to the Brazilian woods: canela-sassafrás, balsamo, and amburana. We also describe the utility of the peptidic sensors as a simple strategy for predicting the identity of unknown cachaça extracts when the clustering patterns are compared to those produced by the training set. This research is part of the curriculum in the Supramolecular Sensors Stream, which is one of the specialized research groups in the Freshman Research Initiative at The University of Texas at Austin.[18]

EXPERIMENTAL SECTION

Materials. The colorimetric indicators Chrome Azurol S (CAS) (purity 65%), Bromopyrogallol Red (BPR), and Pyrocatechol Violet (PCV) (purity 100%) were purchased from Sigma-Aldrich (Saint Louis, MO). Nickel chloride hexahydrate (99.70%), copper(II) sulfate (purity 99.20%), and HEPES buffer were purchased from Fisher Scientific (Hampton, NH). All solid phase peptide synthesis materials were purchased from EMD Millipore (Billerica, MA). All cachaça samples used in this investigation, including the training set (Table 1) and blind unknowns, were provided by Dr. Douglas W. Franco and prepared as previously described.[19] In Table 1, the four pure cachaça samples were not extracted with any wood. Oak samples were obtained from different countries in North America and Europe. The numbers associated with the Brazilian woods canela-sassafrás, balsamo, and amburana indicate that the woods were obtained from different regions in the country. All absorbance measurements were recorded using a Spectra Max Plus 384 plate reader (Molecular Device Inc.).

Indicator Displacement Assay of Cachaça. An array of nine peptide-based sensing ensembles was used in this investigation. Each individual receptor was composed of a short, metal-binding peptide, a divalent metal ion, and a colorimetric indicator. Three peptides were used in our investigation: RN8 (WEEHEE), TT2 (WAHEDEFF), and SEL1 (FHFPHHF). The peptides were synthesized on-site as previously described using a Prelude solid phase peptide synthesizer (Protein Technologies, Inc.).[14] The binding ratios of the ensembles have been previously optimized for discrimination of red wine varietals.[14] Table 2 shows the composition of each receptor and the binding ratios used in the assay. All assays were performed in 96-well plates using 50 mM HEPES buffer, pH 7.4 in 50% ethanol as the solvent. Ethanol was used at this concentration to ensure solubility of all assay components. The final cachaça concentration was 5% (v/v) in 50 mM HEPES buffer, pH 7.4. Displacement of the indicators by the cachaça tannins was monitored at 430, 444, and 560 nm, the λ_{max} of

Table 1. Cachaça Samples Used As the Training Set in This Investigation

Cachaça extract	sample number
Pure Cachaça	Pure Cachaça 1
	Pure Cachaça 2
	Pure Cachaça 3
	Pure Cachaça 4
Oaks	Oak 18 (Scottish Oak)
	Oak 20 (American Oak)
	Oak 24 (French Oak)
	Oak 43
	Oak 45
Canela-sassafrás	Canela-sassafrás 14
	Canela-sassafrás 42
	Canela-sassafrás 59
	Canela-sassafrás 60
Balsamo	Balsamo 21
	Balsamo 53
	Balsamo 55
	Balsamo 58
Amburana	Amburana 28
	Amburana 56
	Amburana 57

Table 2. Composition of Individual Receptors Used in This Investigation

assembly	code	binding ratio (Indicator:M^{2+}:Peptide)	indicator concentration (mM)
PCV-Cu^{2+}-SEL1	PCS	1:1:1	0.075
PCV-Cu^{2+}-TT2	PCT	1:1:0.5	0.075
PCV-Cu^{2+}-RN8	PCR	1:1:0.5	0.075
CAS-Cu^{2+}-SEL1	CCS	1:1:0.5	0.060
CAS-Cu^{2+}-TT2	CCT	1:1:0.4	0.060
CAS-Cu^{2+}-RN8	CCR	1:1:0.4	0.060
BPR-Ni^{2+}-SEL1	BNS	1:1:0.75	0.018
BPR-Ni^{2+}-TT2	BNT	1:1:0.4	0.018
BPR-Ni^{2+}-RN8	BNR	1:1:1	0.018

the free Chrome Azurol S (CAS), Pyrocatechol Violet (PCV), and Bromopyrogallol Red (BPR), respectively. All measurements were taken in either four or seven replicates to ensure reproducibility.

Statistical Data Analysis. Linear discriminant analysis (LDA) was used for cachaça fingerprinting and for determining the classification of unknown samples. Data analysis was done using the software XLSTAT (Addinsoft, New York) and *R*.[20] Absorbance values due to wood extracts of cachaça without the sensing ensembles were subtracted from the final absorbance change before the multivariate data analysis was performed. Input for the LDA consisted of a spreadsheet, where observations were organized in rows and variables were listed in columns. The variables are the sensing ensembles (9 in this case) measured at three different wavelengths, the λ_{max} values for the free indicator. This results in a total of 27 variables. The quantitative observations represent the net change of absorbance at each of the 3 wavelengths for each wood extract. All samples were run in 7 replicates. For the example, the LDA input for the training set shown in Table 1 was a 27×140 matrix (3780 data points).

DOI: 10.1021/acssensors.6b00809
ACS Sens. XXXX, XXX, XXX−XXX

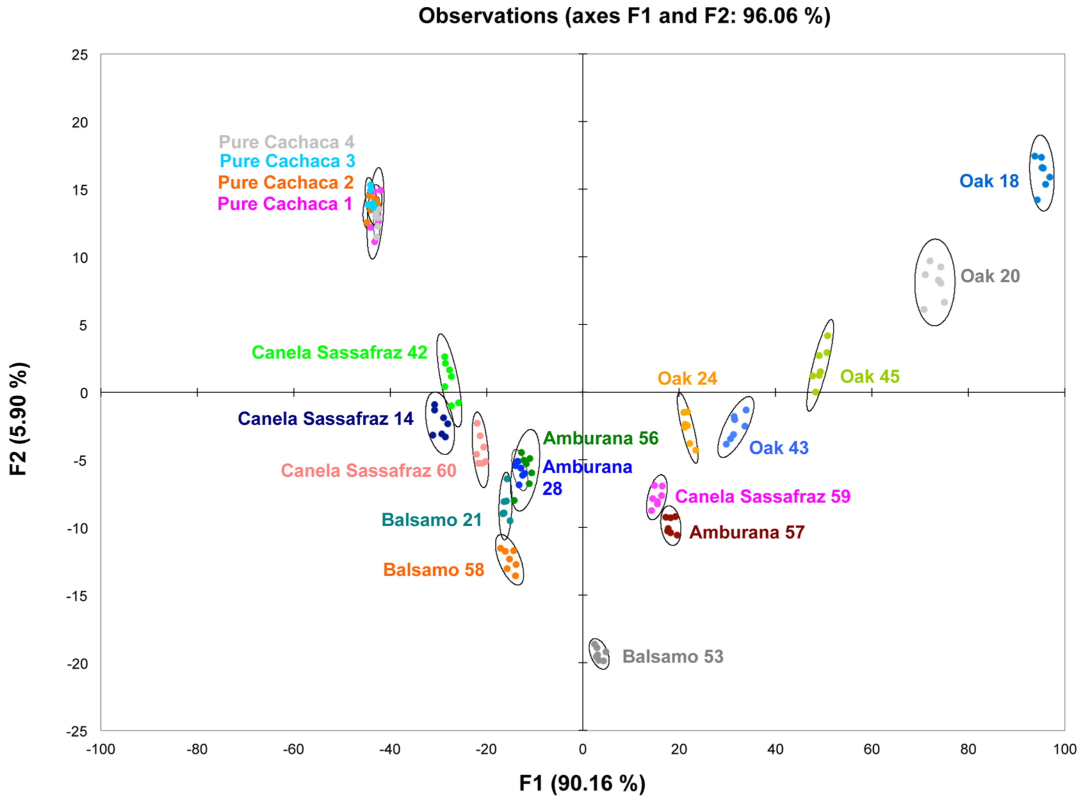


Figure 1. Linear Discriminant Analysis (LDA) score plot of UV−vis responses of the sensing array to cachaça extracts of oaks and Brazilian woods in comparison to pure samples. The numbers for oak samples represent different country of origin, while different numbers associated with Brazilian woods represent different regions within each country.

■ RESULTS AND DISCUSSION

Fingerprinting Cachaça Wood Extracts. We have previously demonstrated the application of a multicomponent, cross reactive peptide-based sensing array to differentiate red wine varietals,[14] to distinguish Cabernet Sauvignon wines based on grapes harvest time[16] and, more recently, to predict the composition of red wine blends.[17] The classification is derived from differential responses of a peptide-based sensing array to tannin composition. In this paper, we describe a new application to differentiate cachaça wood extracts and to predict the identity of unknown samples. A combinatorial library of nine sensing ensembles was used to differentiate sixteen wood extracts in addition to four "pure" samples that had not been extracted with any type of wood, also known as white cachaça. The sensing array contained combinations of three synthetic short peptides: WAHEDEFF (TT2), WEEHEE (RN8), and FHFPHHF (SEL1), as well as three combinations of colorimetric indicators and divalent metal ions. The indicator-metal combinations used in this investigation are Pyrocatechol Violet (PCV)-Cu^{2+}, Chrome Azurol S (CAS)-Cu^{2+}, and Bromopyrogallol Red (BPR)-Ni^{2+}. The color change caused by displacement of the indicator by the tannins released from the wood was used to measure the responses of the array to the wood extracts. The sensing ensembles have been previously tested with tannin concentrations ranging from 0.02 mM to 0.12 mM.[14] The final cachaça concentration was 5% (v/v), which was determined to yield measurable absorbance changes without interference of the unreacted sample (data not shown). All assays were performed in four or seven replicates to ensure precision of measurements. After subtraction of the cachaça absorbance, the data was analyzed by Linear Discriminant Analysis (LDA). Figure 1 shows the resulting canonical variate plots for the training set assay. Numbers of the oak samples indicate the country of origin (Table 1), while numbers of the Brazilian woods (balsamo, canela-sassafrás, and amburana) indicate different regions within the country.

As shown in Figure 1, samples were primarily differentiated with most of the variance explained on the first two dimensions: F1and F2, which had values of 90.16% and 5.90%, respectively. Our previous analysis of responses to this array indicated that differentiation on the first dimension is primarily due to total polyphenols (tannin) concentration.[14] The pure (white) cachaça samples were distinctly differentiated from all other varietals, indicating very low level or lack of polyphenols. All oak samples were clustered in relatively close proximity to each other, yet clearly differentiated from one another. In fact, all oak samples had a slight yellow color, while the pure samples were colorless. Brazilian woods were separated from oaks and from pure cachaça. This pattern is consistent with previous findings on the flavonoid content of cachaça extracted with oak and Brazilian woods. Quantification of phenolic content in cachaça extracts showed higher levels of flavonoids in the oak samples compared to Brazilian woods.[21] The sensing array used in this investigation is composed of cross-reactive sensors that have different binding affinities and, therefore, different sensitivities to the polyphenols present in cachaça. The sensors are neither highly sensitive nor specific to any of the analytes. In other words, individual sensors respond differentially to each analyte, which allows for the discrimination of the complex mixture without the need to identify or characterize each component.[13]

DOI: 10.1021/acssensors.6b00809
ACS Sens. XXXX, XXX, XXX−XXX

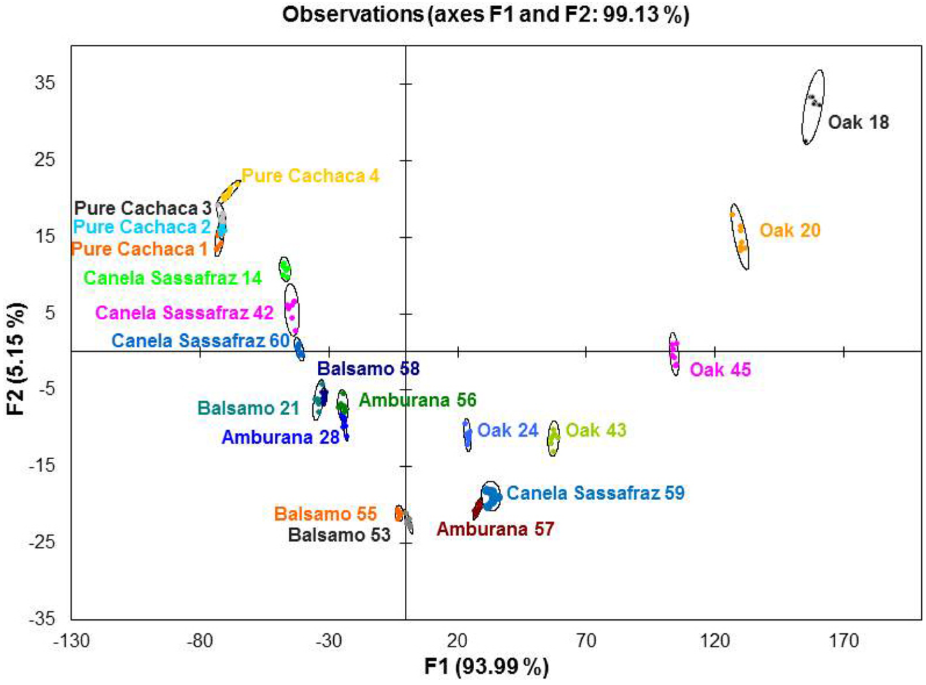


Figure 2. Linear Discriminant Analysis (LDA) score plot for the second screen of cachaça wood extracts training set. The numbers for oak samples represents different countries of origin, while different numbers associated with Brazilian woods represent different regions within each country.

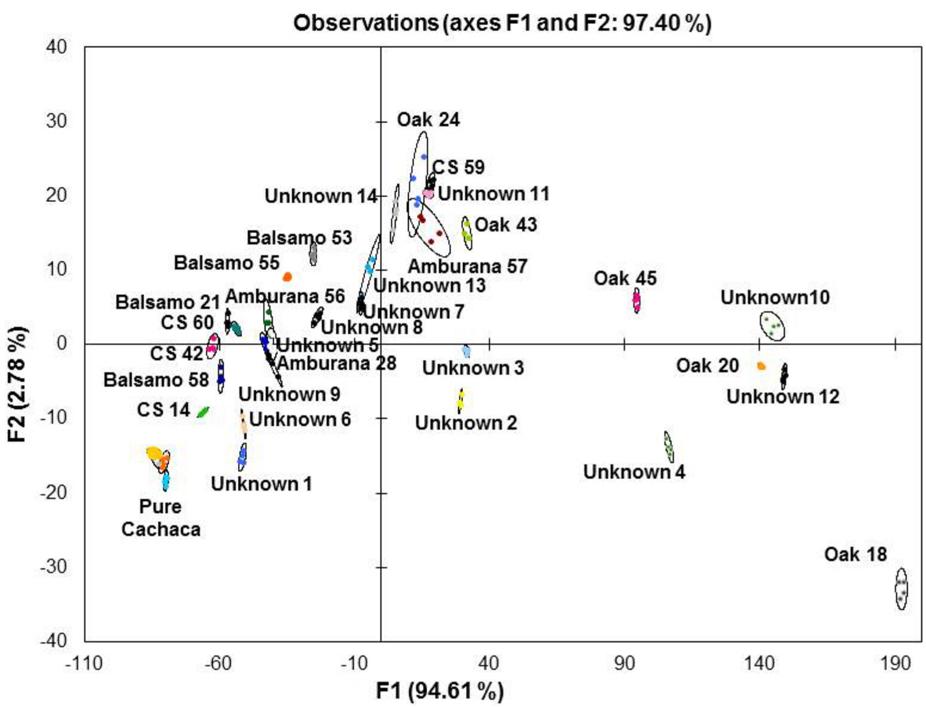


Figure 3. Linear Discriminant Analysis (LDA) score plot for the responses of cachaça wood extracts training set and 14 unknown samples (labeled 1−14). Unknowns were identified based on the clustering pattern in relation to the training set. For simplicity, canela-sassafrás is referred to as "CS".

More interestingly, our data analysis showed that samples extracted with the same wood species grown in different regions in Brazil were also differentiated. For example, amburana 57 was clustered away from amburana 28 and 56, which seem to be more related. Similarly, balsamo 21 was clustered close, yet distinctly separate from balsamo 53 and 58. It was difficult to differentiate between amburana 28 and 56 in this assay. One sample (balsamo 55) was taken out of this analysis because it appeared to be an outlier.

To ensure reproducibility of the classification pattern, the training set screen was independently repeated by a second cohort of students in the FRI Supramolecular Sensors class. The LDA score plot for the assay is shown in Figure 2. The previously missing balsamo 55 extract was included in this assay. As shown in Figure 2, the wood classification pattern was highly reproducible and consistent with the analysis shown in Figure 1. Oak extracts, which contain relatively higher levels of tannins, were clustered farther away from the pure samples.

DOI: 10.1021/acssensors.6b00809
ACS Sens. XXXX, XXX, XXX−XXX

Once again, the assay could not distinctly discriminate between amburana 28 and 56. Canela-sassafrás and amburana 57 appear to have a relatively similar fingerprint to that of oak compared to other Brazilian woods. Given the structure of the FRI class, each year there is a new cohort of students conducting research. It is noteworthy to mention that the second training set screen (Figure 2) was performed one year after completion of the first analysis (Figure 1). This indicates that the sensing array is highly predicable in its response to different cachaça extracts.

An LDA analysis based solely on wood type, regardless of the geographical region, was done using the absorbance measurements obtained from the training set assay (Supporting Information, Figure S1). This analysis revealed that variances within the same wood type are more pronounced than variances between woods. While oaks were clearly discriminated from Brazilian woods, there was a significant overlap between amburana and canela-sassafrás. More defined discrimination patterns were obtained when woods were differentiated based on both type and geographical region.

Identification of Unknown Wood Extracts. The next phase of this investigation focused on using the peptide-based sensing array to determine the identity of blind unknown wood extracts of cachaça for forensic applications. This would be tremendously useful in detecting illegal logging of endangered species[22] and in ensuring authenticity of stored and aged cachaça. Fourteen unknown samples were provided by Dr. Franco's research group at the University of São Paulo and were tested concurrently with the training set as described above. Figure 3 shows the LDA score plot of the training set and the unknowns with 97.40% of the variance explained by the first two dimensions. Classification of the unknowns was predicted based on the clustering pattern of the unknowns relative to the known samples. The correct identity of the samples was not revealed at the time of the assay to avoid any bias in assigning wood types.

Table 3 summarizes the results of our predictions and shows the correct identity of all unknown samples as revealed after completion of the LDA analysis. Two samples were correctly identified as oak: unknown 2 and unknown 4. Unknown 6 was also correctly predicted to be canela-sassafrás. However, the vast majority of the unknowns were incorrectly assigned. Yet, it was then revealed to us that several unknowns were not part of the original training set that was used to "calibrate" the sensing array (amendoim, jequitiba rosa, and castanheira). Therefore, it would not have been possible to correctly predict their identity anyway. Importantly though, by investigating the unknown analysis in greater details, some interesting similarities were found. Unknowns 7 and 13 were repeatedly predicted to be balsamo, while the correct identity is amendoim. In a previous investigation intended to identify Brazilian woods as an alternative to oak in aging cachaça, total phenols measured by UV–vis absorbance values were used to classify eight different Brazilian woods including balsamo and amendoim. Interestingly, Principal Component Analysis (PCA) of the data showed that balsamo and amendoim had similar phenolic content as indicated by the PCA classification pattern, which is consistent with our predictions above.[5]

Table 3. Summary of Predictions of Unknown Wood Extracts and Correct Identification of Each Sample

unknown	prediction	correct identification
1	Canela-sassafrás	Amburana
2	Oak	Oak
3	Oak	Amburana
4	Oak	Oak
5	Amburana 28	Canela-sassafrás
6	Canela-sassafrás 14	Canela-sassafrás
7	Balsamo 53	Amendoim
8	Amburana 56	Balsamo
9	Amburana	Jequitiba rosa
10	Oak 45	Castanheira
11	Canela-sassafrás	Jequitiba rosa
12	Oak 20	Castanheira
13	Balsamo 55	Amendoim
14	Oak 24	Balsamo

Since several unknown samples were not represented in the training set, Franco's research group provided a new set of unknowns that matched the training set to retest the array's efficiency in predicting their identities. In this assay, we anonymously included four of the training set samples as unknowns to examine the effect of sample preparation on the accuracy of predicting wood classification (unknowns 17–20). The LDA score plot of the unknown extracts and the training set is shown in Figure 4. The first two dimensions contributed to the majority of the variance with an F1 value of 90.80% and an F2 value of 5.73%. The clustering pattern of the training set in the presence of the unknowns was consistently reproducible when compared to the analysis of the training set shown in Figures 1 and 2. After removal of all samples that were not part of the training set, six out of ten samples were correctly assigned as summarized in Table 4.

Unknowns 4 and 7 were correctly assigned as balsamo, while unknowns 5 and 6 were classified as oak. Similarly, unknowns 8 and 11 were correctly assigned as canela-sassafrás and amburana. This represents a 60% success rate with no bias against a specific wood species. Unknowns 1 and 2 were clustered between balsamo and amburana; therefore, they were not decisively assigned to either species. Furthermore, it was interesting to see that all four samples anonymously added to the set as unknown were identified correctly. This corresponds to 100% success rate for samples that were extracted and prepared at the same time as the array was created. It is worth mentioning that the timeline for this project spanned a period of three years and the unknown samples were prepared at different times compared to the training set. This may contribute to the variability of observations.

Overall, our current approach of using peptidic sensors for differentiation of polyphenols can potentially provide a forensic tool to classify different species of wood extracts. Additionally, it provides a platform to correlate responses of the array to sensory properties of Brazilian cachaça.[23] Since cachaça is traditionally aged in oak casks, which are not locally available in Brazil, our approach could potentially contribute to identifying Brazilian woods for producing cachaça with similar taste characteristics to that aged in oak barrels. In addition to the applications related to cachaça, the sensors can also be used to classify oak species.

Another important aspect of our findings is the ability of the sensing array to reproducibly differentiate between different oak extracts. Oak is the most commonly used and preferred wood for wine cooperage and the source of oak is crucial to winemakers because of the genetic diversities of oak species that grow in different regions.[24] As we demonstrated in this investigation, this peptide-based sensing array can easily be

DOI: 10.1021/acssensors.6b00809
ACS Sens. XXXX, XXX, XXX–XXX

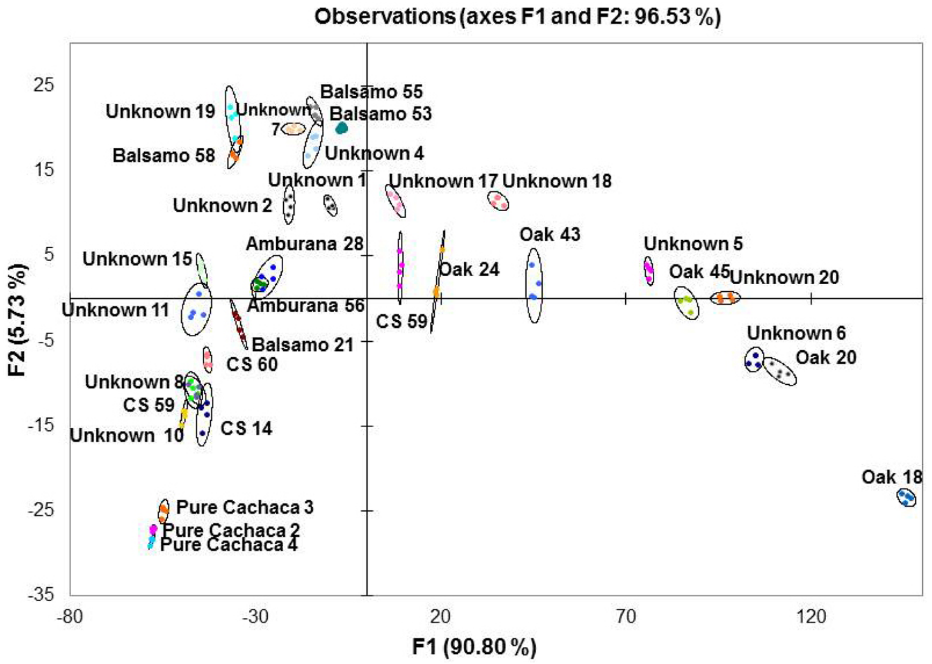


Figure 4. Linear Discriminant Analysis (LDA) score plot for ten unknown cachaça wood extracts. Four extracts from the training set were anonymously included in this assay as unknowns (17−20). For simplicity, canela-sassafrás is referred to as "CS".

Table 4. Predicted Classification of the Second Set of Cachaça Unknowns after Removal of Samples Not Included in the Training Set

unknown	prediction	correct identification
1	Balsamo or Amburana	Amburana 25
2	Balsamo or Amburana	Amburana 28
4	Balsamo	Balsamo 17
5	Oak	Oak 26
6	Oak 20	Oak 23
7	Balsamo	Balsamo 16
8	Canela-sassafrás	Canela-sassafrás 42
10	Canela-sassafrás	Campanari Cachaça Traditional Oak
11	Amburana	Campanari Cachaça Amburana
15	Amburana	Canela-sassafrás 2011
17	Canela-sassafrás	Canela-sassafrás 59
18	Oak 43	Oak 43
19	Balsamo 58	Balsamo 58
20	Oak 20	Oak 20

adapted to differentiate oak species for wine authenticity and quality control. There is an increasing demand to identify more cost efficient alternatives to French oak, which is currently the preferred type of oak for aging red wine.[25,26]

■ CONCLUSIONS

In the present study, we have demonstrated a new application for cross-reactive, multicomponent receptors composed of a short peptide, a divalent metal ion, and a colorimetric indicator in fingerprinting cachaça extracts of oak and Brazilian woods based on their tannin composition. The discrimination pattern was reproducible for oaks obtained from different countries and for regional Brazilian woods. Furthermore, the assay was used to classify unknown wood extracts based on their clustering pattern with 60−100% success rates. Our findings provide a foundation for developing robust and inexpensive multi-component sensors for post-processing wood identification, which can be broadly used for quality control and forensic applications.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acssensors.6b00809.

Linear Discriminant Analysis (LDA) of cachaça extracts based on wood type only and regardless of the geographical region (PDF)

■ AUTHOR INFORMATION

Corresponding Author

*E-mail: anslyn@austin.utexas.edu. Tel.: +1-512-471-0068. Fax: +1-512-471-6835.

ORCID

Eman Ghanem: 0000-0002-7024-5770

Eric V. Anslyn: 0000-0002-5137-8797

Present Address

#Sigma Xi, The Scientific Research Honor Society, P.O. Box 13975, Research Triangle Park, NC 27709

Author Contributions

¶The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript. These authors contributed equally.

Funding

Funding for this research was provided by the Freshman Research Initiative of the College of Natural Sciences at The University of Texas at Austin. We also acknowledge the financial support of the National Science Foundation (Grant CHE-0716049), the Howard Hughes Medical Institute (Grant 52006985) and the Welch Regent Chair (Grant F-0046).

Notes

The authors declare no competing financial interest.

DOI: 10.1021/acssensors.6b00809
ACS Sens. XXXX, XXX, XXX−XXX

ACKNOWLEDGMENTS

We would like to thank the following students in the FRI Supramolecular Sensors Stream for their contribution to this research: Benjamin Chang, Andrea Navarro, Maxwell Ritzer, and Mathew Stephen. We would also like to thank the FRI peer mentors: Syman Lau, Jevons Wang, and Shunan Zhu for assisting the students.

DEDICATION

△Authors would like to dedicate this manuscript to the memory of Dr. Douglas W. Franco (1945−2016).

ABBREVIATIONS

CAS, Chrome Azurol S; BPR, Bromopyrogallol Red; PCV, Pyrocatechol Violet; HEPES, 4-(2-hydroxyethyl)-1 piperazineethanesulfonic acid

REFERENCES

(1) Faria, J. B.; Franco, D. W.; Piggott, J. R. The quality challenge: cachaça for export in the 21st century. In *Distilled spirits: tradition and innovation*, Bryce, J. H.; Steward, G. G., Eds.; Nottingham University Press: Nottingham, England, 2004; pp 215−221.

(2) Reche, R. V.; Leite Neto, A. F.; Da Silva, A. A.; Galinaro, C. A.; De Osti, R. Z.; Franco, D. W. Influence of type of distillation apparatus on chemical profiles of Brazilian cachaças. *J. Agric. Food Chem.* **2007**, *55*, 6603−6608.

(3) Maga, J. A. The contribution of wood to the flavor of alcoholic beverages. *Food Rev. Int.* **1989**, *5* (1), 39−99.

(4) Aquino, F. W. B.; Rodrigues, S.; Nascimento, R. F.; Casimiro, A. R. S. Phenolic compounds in imburana (Amburana cearensis) powder extracts. *Eur. Food Res. Technol.* **2005**, *221*, 739−745.

(5) Faria, J. B.; Cardello, H. M. A. B.; Boscolo, M.; Isique, W. D.; Odello, L.; Franco, D. W. Evaluation of Brazilian woods as an alternative to oak for cachaça aging. *Eur. Food Res. Technol.* **2003**, *218*, 83−87.

(6) Da Silva, A. A.; Do Nascimento, E. S.; Cardoso, D. R.; Franco, D. W. Coumarins and phenolic fingerprints of oak and Brazilian woods extracted by sugarcane spirit. *J. Sep. Sci.* **2009**, *32*, 3681−3691.

(7) Bortoletto, A. M.; Alcarde, A. R. Congeners in sugar cane spirits aged in casks of different woods. *Food Chem.* **2013**, *139*, 695−701.

(8) De Souza, P. P.; Siebald, H. G. L.; Augusti, D. V.; Neto, W. B.; Amorim, V. M.; Catharino, R. R.; Eberlin, M. N.; Augusti, R. Electrospray ionization mass spectrometry fingerprinting of Brazilian artisan Cachaça aged in different wood casks. *J. Agric. Food Chem.* **2007**, *55*, 2094−2102.

(9) Core, H. A.; Côté, W. A.; Day, A. C. *Wood structure and identification*, 2nd ed.; Syracuse Wood Science Series 6; Syracuse University Press: Syracuse, NY,1979.

(10) Cooper, P. A.; Jeremic, D.; Radivojevic, S.; Ung, Y. T.; Leblon, B. Potential of near-infrared spectroscopy to characterize wood products. *Can. J. For. Res.* **2011**, *41*, 2150−2157.

(11) Lawrence, A. H.; Barbour, R. J.; Sutcliffe, R. Identification of wood species by ion mobility spectrometry. *Anal. Chem.* **1991**, *63*, 1217−1221.

(12) Cordeiro, J. R.; Martinez, M. I. V.; Li, R. W.C.; Cardoso, A. P.; Nunes, L. C.; Krug, F. J.; Paixão, T. R. L. C.; Nomura, C. S.; Gruber, J. Identification of four wood species by an electronic nose and by LIBS. *Int. J. Electrochem.* **2012**, *2012*, 1−5.

(13) Umali, A. P.; Anslyn, E. V. A General Approach to Differential Sensing Using Synthetic Molecular Receptors. *Curr. Opin. Chem. Biol.* **2010**, *14*, 685−692.

(14) Umali, A. P.; LeBoeuf, S. E.; Newberry, R. W.; Kim, S.; Tran, L.; Rome, W. A.; Tian, T.; Taing, D.; Hong, J.; Kwan, M.; Heymann, H.; Anslyn, E. V. Discrimination of flavonoids and red wine varietals by arrays of differential peptidic sensors. *Chem. Sci.* **2011**, *2*, 439−445.

(15) Nguyen, B. T.; Anslyn, E. V. Indicator Displacement Assays. *Coord. Chem. Rev.* **2006**, *250*, 3118−3127.

(16) Umali, A. P.; Ghanem, E.; Hopfer, H.; Hussain, A.; Kao, Y.; Zabanal, L. G.; Wilkins, B. J.; Hobza, C.; Quach, D. K.; Fredell, M.; Heymann, H.; Anslyn, E. V. Grape and wine sensory attributes correlate with pattern-based discrimination of Cabernet Sauvignon wines by a peptidic sensor array. *Tetrahedron* **2015**, *71*, 3095.

(17) Ghanem, E.; Hopfer, H.; Navarro, A.; Ritzer, M. S.; Mahmood, L.; Fredell, M.; Cubley, A.; Bolen, J.; Fattah, R.; Teasdalea, K.; Lieu, L.; Chua, T.; Marini, F.; Heymann, H.; Anslyn, E. V. Predicting the Composition of Red Wine Blends Using an Array of Multicomponent Peptide-Based Sensors. *Molecules* **2015**, *20*, 9170−9182.

(18) Rodenbusch, S. E.; Hernandez, P. R.; Simmons, S. L.; Dolan, E. L. Early Engagement in Course-Based Research Increases Graduation Rates and Completion of Science, Engineering, and Mathematics Degrees. *CBE Life Sci. Educ.* **2016**, *15* (2), 1−10.

(19) Da Silva, A. A.; De Keukeleire, D.; Cardoso, D. R.; Franco, D. W. Multivariate analyses of UV-Vis absorption spectral data from cachaça wood extracts: a model to classify aged Brazilian cachaças according to the wood species used. *Anal. Methods* **2012**, *4*, 642−646.

(20) R Core Team (2013). *R: A language and environment for statistical computing*; R Foundation for Statistical Computing; Vienna, Austria; http://www.R-project.org/.

(21) Cardoso, D. R.; Frederiksen, A. M.; Ataide Da Silva, A.; Franco, D. W.; Skibsted, L. H. Sugarcane spirit extracts of oak and Brazilian woods: antioxidant capacity and activity. *Eur. Food Res. Technol.* **2008**, *227*, 1109−1116.

(22) Smith, W. The global problem of illegal logging. *Tropical Forest Update* **2002**, *12* (1), 3−5.

(23) Serafim, F. A. T.; Seixas, F. R. F.; Da Silva, A. A.; Galinaro, C. A.; Nascimento, E. S. P.; Buchviser, S. F.; Odello, L.; Franco, D. W. Correlation between chemical composition and sensory properties of Brazilian sugarcane spirits (Cachaças). *J. Braz. Chem. Soc.* **2013**, *24*, 973−982.

(24) Garde-Cerdán, T.; Ancín-Azpilicueta, C. Review of quality factors on wine aging in oak barrels. *Trends Food Sci. Technol.* **2006**, *17*, 438−447.

(25) Waterhouse, A. L.; Towey, J. P. Oak lactone isomer ration distinguishes between wines fermented in American and French oak barrels. *J. Agric. Food Chem.* **1994**, *42*, 1971−1974.

(26) De Simón, B. F.; Cadahía, E.; Jalocha, J. Volatile Compounds in a Spanish red wine aged in barrels made of Spanish, French and American oak wood. *J. Agric. Food Chem.* **2003**, *51*, 7671−7678.

DOI: 10.1021/acssensors.6b00809
ACS Sens. XXXX, XXX, XXX−XXX