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Application Number: 15913090 Document Date: 03/06/2018

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Street Address
Fish & Richardson P.C.
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60 South Sixth Street
Minneapolis, MN 55402

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612 335 5070 main
877 769 7945 fax

March 6, 2018

Attorney Docket No.: 38767-0015008

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Presented for filing is a new Continuation patent application of:

Inventor(s): RACHEL FRASER, PATRICK O'REILLY BROWN M.D., PH.D., JESSICA KARR, CELESTE HOLZ-SCHIETINGER AND ELYSIA P. COHN

Title: METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES

Enclosed are the following papers, including those required to receive a filing date under 37 CFR § 1.53(b):

	Pages
Application Data Sheet	10
Specification	67
Claims	4
Abstract	1
Drawing(s)	5
Nucleotide and/or amino acid sequence listing (.txt format)	
Declaration	5
Power of Attorney	2

Enclosures:

- Exhibit 1 – Holz-Schietinger Declaration (w/exhibits)
- Exhibit 2 – Cadwallader Declaration (w/exhibits)
- Exhibit 3 – Peterson Declaration (w/exhibits)
- Exhibit 4 – Holz-Schietinger Declaration (Heme Gradient) (w/exhibits)
- Exhibit 5 – Holz-Schietinger Declaration (Low Sulfur) (w/exhibits)
- Exhibit 6 – Davis Declaration (19 July 2017) (w/exhibits)
- Exhibit 7 – Lipman Declaration (HemeAlignment) (w/exhibits)

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March 6, 2018

Basic Filing Fee			$300
Search Fee			$660
Examination Fee			$760
Total Claims (20)	over 20	0 x $100	$0
Independent Claims (2)	over 3	0 x $460	$0
Fee for Multiple Dependent claims			$0
Application size fee for each 50 pages over 100			$0
Total Filing fee			$1720

The filing fee is being paid concurrently via the Electronic Filing System (EFS) by Deposit Account authorization. Please apply any other fees, or credits, to Deposit Account No. 06-1050, referencing the Attorney Docket Number shown above.

I hereby authorize the USPTO to communicate via email in response to email communications from the undersigned and practitioners under Customer No. 26191 in accordance with 37 CFR § 1.33 and 37 CFR § 1.34 concerning any subject matter of this application. I understand that a copy of these communications will be made of record in the application file. (MPEP § 502.03).

If this application is considered to be incomplete, or if a telephone conference would otherwise be helpful, call the undersigned at (612) 335-5070.

Send all correspondence to:

26191
Customer Number

Respectfully submitted,

/M. Angela Parsons/

M. Angela Parsons, Ph.D.
Registration No. 44,282

Enclosures
MAP/jxw

61644798.doc

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

The application data sheet is part of the provisional or nonprovisional application for which it is being submitted. The following form contains the bibliographic data arranged in a format specified by the United States Patent and Trademark Office as outlined in 37 CFR 1.76.
This document may be completed electronically and submitted to the Office in electronic format using the Electronic Filing System (EFS) or the document may be printed and included in a paper filed application.

Secrecy Order 37 CFR 5.2:

☐ Portions or all of the application associated with this Application Data Sheet may fall under a Secrecy Order pursuant to 37 CFR 5.2 (Paper filers only. Applications that fall under Secrecy Order may not be filed electronically.)

Inventor Information:

Inventor 1 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Rachel		Fraser	

Residence Information (Select One) ● US Residency Non US Residency Active US Military Service

City	State/Province	Country of Residence
San Francisco	CA	US

Mailing Address of Inventor:

Address 1	123 Connecticut Street
Address 2	
City	San Francisco
State/Province	CA
Postal Code	94107
Country	US

Inventor 2 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
Dr.	Patrick	O'Reilly	Brown	

Residence Information (Select One) ◉ US Residency Non US Residency Active US Military Service

City	State/Province	Country of Residence
Stanford	CA	US

Mailing Address of Inventor:

Address 1	76 Peter Coutts Circle
Address 2	
City	Stanford
State/Province	CA
Postal Code	94305
Country	US

Inventor 3 Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Jessica		Karr	

Residence Information (Select One) ◉ US Residency Non US Residency Active US Military Service

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

City	San Francisco	State/Province	CA	Country of Residence	US

Mailing Address of Inventor:

Address 1	456 Kansas Street
Address 2	

City	San Francisco	State/Province	CA
Postal Code	94107	Country	US

Inventor 4 — Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Celeste		Holz-Schietinger	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	East Palo Alto	State/Province	CA	Country of Residence	US

Mailing Address of Inventor:

Address 1	1159 Sage Street
Address 2	

City	East Palo Alto	State/Province	CA
Postal Code	94303	Country	US

Inventor 5 — Remove

Legal Name

Prefix	Given Name	Middle Name	Family Name	Suffix
	Elysia		Cohn	

Residence Information (Select One) ◉ US Residency ○ Non US Residency ○ Active US Military Service

City	Sunnyvale	State/Province	CA	Country of Residence	US

Mailing Address of Inventor:

Address 1	1351 Navarro Drive
Address 2	

City	Sunnyvale	State/Province	CA
Postal Code	94087	Country	US

All Inventors Must Be Listed - Additional Inventor Information blocks may be generated within this form by selecting the **Add** button. Add

Correspondence Information:

Enter either Customer Number or complete the Correspondence Information section below.
For further information see 37 CFR 1.33(a).

☐ **An Address is being provided for the correspondence Information of this application.**

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

Customer Number	26191		
Email Address	apsi@fr.com	Add Email	Remove Email

Application Information:

Title of the Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES		
Attorney Docket Number	38767-0015008	**Small Entity Status Claimed**	☐
Application Type	Nonprovisional		
Subject Matter	Utility		
Total Number of Drawing Sheets (if any)	5	**Suggested Figure for Publication (if any)**	

Filing By Reference:

Only complete this section when filing an application by reference under 35 U.S.C. 111(c) and 37 CFR 1.57(a). Do not complete this section if application papers including a specification and any drawings are being filed. Any domestic benefit or foreign priority information must be provided in the appropriate section(s) below (i.e., "Domestic Benefit/National Stage Information" and "Foreign Priority Information").

For the purposes of a filing date under 37 CFR 1.53(b), the description and any drawings of the present application are replaced by this reference to the previously filed application, subject to conditions and requirements of 37 CFR 1.57(a).

Application number of the previously filed application	Filing date (YYYY-MM-DD)	Intellectual Property Authority or Country

Publication Information:

☐ Request Early Publication (Fee required at time of Request 37 CFR 1.219)

☐ **Request Not to Publish.** I hereby request that the attached application not be published under 35 U.S.C. 122(b) and certify that the invention disclosed in the attached application **has not and will not** be the subject of an application filed in another country, or under a multilateral international agreement, that requires publication at eighteen months after filing.

Representative Information:

Representative information should be provided for all practitioners having a power of attorney in the application. Providing this information in the Application Data Sheet does not constitute a power of attorney in the application (see 37 CFR 1.32).
Either enter Customer Number or complete the Representative Name section below. If both sections are completed the customer Number will be used for the Representative Information during processing.

Please Select One:	● Customer Number	US Patent Practitioner	○ Limited Recognition (37 CFR 11.9)
Customer Number	26191		

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Domestic Benefit/National Stage Information:

This section allows for the applicant to either claim benefit under 35 U.S.C. 119(e), 120, 121, 365(c), or 386(c) or indicate National Stage entry from a PCT application. Providing benefit claim information in the Application Data Sheet constitutes the specific reference required by 35 U.S.C. 119(e) or 120, and 37 CFR 1.78.
When referring to the current application, please leave the "Application Number" field blank.

Prior Application Status	Pending		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
	Continuation of	15398479	2017-01-04
Prior Application Status	Patented		Remove

Application Number	Continuity Type	Prior Application Number	Filing Date (YYYY-MM-DD)	Patent Number	Issue Date (YYYY-MM-DD)
15398479	Continuation of	14797006	2015-07-10	9700067	2017-07-11

Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
14797006	Continuation of	PCTUS2014011347	2014-01-13
Prior Application Status	Pending		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
PCTUS2014011347	Continuation of	13941211	2013-07-12
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
PCTUS2014011347	Claims benefit of provisional	61908634	2013-11-25
Prior Application Status	Expired		Remove
Application Number	Continuity Type	Prior Application Number	Filing or 371(c) Date (YYYY-MM-DD)
PCTUS2014011347	Claims benefit of provisional	61751816	2013-01-11

Additional Domestic Benefit/National Stage Data may be generated within this form by selecting the **Add** button. Add

Foreign Priority Information:

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

This section allows for the applicant to claim priority to a foreign application. Providing this information in the application data sheet constitutes the claim for priority as required by 35 U.S.C. 119(b) and 37 CFR 1.55. When priority is claimed to a foreign application that is eligible for retrieval under the priority document exchange program (PDX)[i] the information will be used by the Office to automatically attempt retrieval pursuant to 37 CFR 1.55(i)(1) and (2). Under the PDX program, applicant bears the ultimate responsibility for ensuring that a copy of the foreign application is received by the Office from the participating foreign intellectual property office, or a certified copy of the foreign priority application is filed, within the time period specified in 37 CFR 1.55(g)(1).

Remove

Application Number	Country[i]	Filing Date (YYYY-MM-DD)	Access Code[i] (if applicable)

Additional Foreign Priority Data may be generated within this form by selecting the **Add** button. Add

Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications

☐ This application (1) claims priority to or the benefit of an application filed before March 16, 2013 and (2) also contains, or contained at any time, a claim to a claimed invention that has an effective filing date on or after March 16, 2013.
NOTE: By providing this statement under 37 CFR 1.55 or 1.78, this application, with a filing date on or after March 16, 2013, will be examined under the first inventor to file provisions of the AIA.

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

Authorization or Opt-Out of Authorization to Permit Access:

When this Application Data Sheet is properly signed and filed with the application, applicant has provided written authority to permit a participating foreign intellectual property (IP) office access to the instant application-as-filed (see paragraph A in subsection 1 below) and the European Patent Office (EPO) access to any search results from the instant application (see paragraph B in subsection 1 below).

Should applicant choose not to provide an authorization identified in subsection 1 below, applicant **must opt-out** of the authorization by checking the corresponding box A or B or both in subsection 2 below.

NOTE: This section of the Application Data Sheet is **ONLY** reviewed and processed with the **INITIAL** filing of an application. After the initial filing of an application, an Application Data Sheet cannot be used to provide or rescind authorization for access by a foreign IP office(s). Instead, Form PTO/SB/39 or PTO/SB/69 must be used as appropriate.

1. Authorization to Permit Access by a Foreign Intellectual Property Office(s)

A. Priority Document Exchange (PDX) - Unless box A in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the European Patent Office (EPO), the Japan Patent Office (JPO), the Korean Intellectual Property Office (KIPO), the State Intellectual Property Office of the People's Republic of China (SIPO), the World Intellectual Property Organization (WIPO), and any other foreign intellectual property office participating with the USPTO in a bilateral or multilateral priority document exchange agreement in which a foreign application claiming priority to the instant patent application is filed, access to: (1) the instant patent application-as-filed and its related bibliographic data, (2) any foreign or domestic application to which priority or benefit is claimed by the instant application and its related bibliographic data, and (3) the date of filing of this Authorization. See 37 CFR 1.14(h)(1).

B. Search Results from U.S. Application to EPO - Unless box B in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the EPO access to the bibliographic data and search results from the instant patent application when a European patent application claiming priority to the instant patent application is filed. See 37 CFR 1.14(h)(2).

The applicant is reminded that the EPO's Rule 141(1) EPC (European Patent Convention) requires applicants to submit a copy of search results from the instant application without delay in a European patent application that claims priority to the instant application.

2. Opt-Out of Authorizations to Permit Access by a Foreign Intellectual Property Office(s)

☐ A. Applicant **DOES NOT** authorize the USPTO to permit a participating foreign IP office access to the instant application-as-filed. If this box is checked, the USPTO will not be providing a participating foreign IP office with any documents and information identified in subsection 1A above.

☐ B. Applicant **DOES NOT** authorize the USPTO to transmit to the EPO any search results from the instant patent application. If this box is checked, the USPTO will not be providing the EPO with search results from the instant application.

NOTE: Once the application has published or is otherwise publicly available, the USPTO may provide access to the application in accordance with 37 CFR 1.14.

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

Applicant Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

Applicant 1 Remove

If the applicant is the inventor (or the remaining joint inventor or inventors under 37 CFR 1.45), this section should not be completed. The information to be provided in this section is the name and address of the legal representative who is the applicant under 37 CFR 1.43; or the name and address of the assignee, person to whom the inventor is under an obligation to assign the invention, or person who otherwise shows sufficient proprietary interest in the matter who is the applicant under 37 CFR 1.46. If the applicant is an applicant under 37 CFR 1.46 (assignee, person to whom the inventor is obligated to assign, or person who otherwise shows sufficient proprietary interest) together with one or more joint inventors, then the joint inventor or inventors who are also the applicant should be identified in this section.

Clear

● Assignee	Legal Representative under 35 U.S.C. 117	Joint Inventor
Person to whom the inventor is obligated to assign.	Person who shows sufficient proprietary interest	

If applicant is the legal representative, indicate the authority to file the patent application, the inventor is:

Name of the Deceased or Legally Incapacitated Inventor:

If the Applicant is an Organization check here. ☒

Organization Name	Impossible Foods Inc.		
Mailing Address Information For Applicant:			
Address 1	525 Chesapeake Drive		
Address 2			
City	Redwood City	**State/Province**	CA
Country	US	Postal Code	94063
Phone Number		Fax Number	
Email Address			

Additional Applicant Data may be generated within this form by selecting the Add button. Add

Assignee Information including Non-Applicant Assignee Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

Assignee 1

Complete this section if assignee information, including non-applicant assignee information, is desired to be included on the patent application publication. An assignee-applicant identified in the "Applicant Information" section will appear on the patent application publication as an applicant. For an assignee-applicant, complete this section only if identification as an assignee is also desired on the patent application publication.

Remove

If the Assignee or Non-Applicant Assignee is an Organization check here. ☐

Prefix	**Given Name**	Middle Name	**Family Name**	Suffix

Mailing Address Information For Assignee including Non-Applicant Assignee:

Address 1			
Address 2			
City		**State/Province**	
Country i		Postal Code	
Phone Number		Fax Number	
Email Address			

Additional Assignee or Non-Applicant Assignee Data may be generated within this form by selecting the Add button. Add

Signature:

Remove

NOTE: This Application Data Sheet must be signed in accordance with 37 CFR 1.33(b). **However, if this Application Data Sheet is submitted with the INITIAL filing of the application and either box A or B is not checked in subsection 2 of the "Authorization or Opt-Out of Authorization to Permit Access" section, then this form must also be signed in accordance with 37 CFR 1.14(c).**

This Application Data Sheet **must** be signed by a patent practitioner if one or more of the applicants is a **juristic entity** (e.g., corporation or association). If the applicant is two or more joint inventors, this form must be signed by a patent practitioner, **all** joint inventors who are the applicant, or one or more joint inventor-applicants who have been given power of attorney (e.g., see USPTO Form PTO/AIA/81) on behalf of **all** joint inventor-applicants.

See 37 CFR 1.4(d) for the manner of making signatures and certifications.

Signature	/M. Angela Parsons/			Date (YYYY-MM-DD)	2018-03-06
First Name	M. Angela	Last Name	Parsons	Registration Number	44282

Additional Signature may be generated within this form by selecting the Add button. Add

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Application Data Sheet 37 CFR 1.76	Attorney Docket Number	38767-0015008
	Application Number	
Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES	

This collection of information is required by 37 CFR 1.76. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 23 minutes to complete, including gathering, preparing, and submitting the completed application data sheet form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1 The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether the Freedom of Information Act requires disclosure of these records.

2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.

3 A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.

4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).

5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent CooperationTreaty.

6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).

7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.

8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspections or an issued patent.

9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES

CROSS-REFERENCE TO RELATED APPLICATIONS

This application is a Continuation of U.S. Patent Application no. 15/398,479 filed January 4, 2017, which is a Continuation of U.S. Patent Application No. 14/797,006 filed July 10, 2015, which is a Continuation of PCT/US2014/011347 which is a Continuation of and claims priority to U.S. Application Serial No. 13/941,211, filed July 12, 2013, U.S. Application Serial No. 61/908,634, filed November 25, 2013, and to U.S. Application Serial No. 61/751,816, filed January 11, 2013, and is related to the following co-pending patent applications: Application Serial No. PCT/US12/46560; Application Serial No PCT/US12/46552; Application Serial No. 61,876,676, filed September 11, 2013; and Application Serial No. 61/751,818, filed January 11, 2013, all of which are incorporated herein by reference.

TECHNICAL FIELD

This invention relates to food products and more particularly, to food products that include a highly conjugated heterocyclic ring complexed to iron such as a heme-cofactor and one or more flavor precursor molecules.

BACKGROUND

Food is any substance that is either eaten or drunk by any animal, including humans, for nutrition or pleasure. It is usually of plant or animal origin, and can contain essential nutrients, such as carbohydrates, fats, proteins, vitamins, or minerals. The substance is ingested by an organism and assimilated by the organism's cells in an effort to produce energy, maintain life, or stimulate growth.

Food typically has its origin in a photosynthetic organism, such as a plant. Some food is obtained directly from plants, but even animals that are used as food sources are raised by feeding them food which is typically derived from plants.

In most cases, the plant or animal food source is fractionated into a variety of different portions, depending upon the purpose of the food. Often, certain portions of the plant, such as the seeds or fruits, are more highly prized by humans than others and these are selected for human consumption, while other less desirable portions, such as the stalks of grasses, are typically used for feeding animals.

Current plant-based meat substitutes have largely failed to cause a shift to a vegetarian diet. Meat substitute compositions are typically extruded soy/grain mixtures which largely fail to replicate the experience of cooking and eating meat. Common limitations of plant-based meat substitute products are a texture and mouth-feel that are more homogenous than that of equivalent meat products. Furthermore, as these products must largely be sold pre-cooked, with artificial flavors and aromas pre-incorporated, they fail to replicate the aromas, flavors, and other key features, such as texture and mouth-feel, associated with cooking or cooked meat. As a result, these products appeal largely to a limited consumer base that is already committed to vegetarianism/veganism, but have failed to appeal to the larger consumer segment accustomed to eating meat. It would be useful to have improved plant-based meat substitutes which better replicate the aromas and flavors of meat, particularly during and/or after cooking.

SUMMARY

Provided herein are methods and compositions for modulating the flavor and/or aroma profile of consumable food products, including animal- or non-animal (e.g., plant) based food products, or mixtures of animal- and non-animal-based food products. In some embodiments, the methods and compositions are useful for modulating the flavor and/or aroma profile of a consumable food product during and/or after the cooking process. In some embodiments, the methods and compositions are used to generate one or more chemical compounds that modulate the flavor and/or aroma profile of the consumable food product during and/or after the cooking process.

As provided herein, and without being bound by theory, certain characteristic meaty flavors and/or aromas (e.g., beefy, bacony, umami, savory, bloody, brothy, gravy, metallic, bouillon-like; see Tables 2, 7, and 11), including one or more specific chemical

compounds associated with the same (see Tables 3, 8, 9, 12, 14, 16, or 17), are believed to be produced during the cooking process of a consumable food product by chemical reaction of one or more flavor precursor molecules or compositions catalyzed by the presence of a highly conjugated heterocyclic ring complexed to an iron ion (e.g., a heme moiety; or a porphyrin; a porphyrinogen; a corrin; a corrinoid; a chlorin; a bacteriochorophyll; a corphin; a chlorophyllin; a bacteriochlorin; or an isobacteriochlorin moiety complexed to an iron ion). Such highly conjugated heterocycylic moieties include heterocyclic aromatic rings composed of one or more (2, 3, or 4 more) pyrrole, pyrrole-like, and/or pyrroline subunits. The highly conjugated heterocyclic ring complexed to an iron ion is referred to herein as an iron complex. In some embodiments, the heme moiety can be a heme cofactor such as a heme moiety bound to a protein; a heme moiety bound to a non-proteinaceous polymer; a heme moiety bound to a solid support; or a heme moiety encapsulated in a liposome. In some embodiments, the flavors and/or aromas are not generated in the absence of the iron complex (e.g., in the absence of a ferrous chlorin) or are not generated in the absence of a heme-cofactor (e.g., in the absence of a heme-containing protein). Accordingly, as described herein, the iron complexes such as isolated chlorin-iron complexes or heme-cofactors (e.g., heme-containing proteins) can be used to generate meaty flavors and/or aromas in a variety of food products, such as during the cooking process.

Combining one or more iron complexes such as a heme-cofactor (e.g., a heme-containing protein, including, for example a plant-derived heme protein such as a plant leghemoglobin (legH)), with one or more flavor precursor molecules or compositions (see, e.g., Table 1 or Table 13) can generate or provide a range of savory and meaty aromas and tastes (see, e.g., Tables 2, 7, and/or 11) in a cooked consumable food product. Flavor precursor molecules or compositions can be added to the uncooked food product in purified form and/or can be derived from ingredients in the uncooked consumable food product that contain and/or are enriched with one or more of the particular flavor precursors or compositions, including, for example, yeast extract, vegetable oil, corn oil, soybean oil, palm fruit oil, palm kernel oil, safflower oil, flaxseed oil, rice bran oil, cottonseed oil, olive oil, canola oil, sunflower oil, coconut oil, mango oil, or an algal oil.

The resultant flavor and/or aroma profile can be modulated by the type and concentration of the flavor precursors, the pH of the reaction, the length of cooking, the type and amount of iron complex (e.g., a heme cofactor such as a heme-containing protein), the temperature of the reaction, and the amount of water activity in the product, among other factors.

One or more flavor precursor molecules or compositions can be added along with a iron complex (e.g., ferrous chlorophyllin or a heme cofactor such as a heme-containing protein), to an uncooked food product, before and/or during the cooking process, to give the cooked consumable food product a particular meaty taste and smell, for example, the taste and smell of beef, bacon, pork, lamb, or chicken. Consumable food products can be animal or non-animal based (e.g., plant) food products, or combinations of an animal and non-animal based food product. For example, a plant based veggie burger or an animal-based burger, such as a chicken burger, can be modified with the compositions and methods of the present disclosure to result in a burger having a cooked flavor and/or aroma profile that is more meat like, e.g., beef-like, lamb-like, pork-like, turkey-like, duck-like, deer-like, yak-like, bison-like or other desirable meat flavor.

Food products for use in the present disclosure include those that have an iron-complex (e.g., a heme cofactor such as a heme-containing protein), and one or more flavor precursor molecules included therein. The iron-complex such as a heme cofactor (e.g., a heme-containing protein) and the one or more flavor precursor molecules can be homogenously or heterogeneously included in the food products. A heme protein can be isolated and purified prior to inclusion in the food product. Non-limiting examples of consumable food products which can include an iron complex such as a heme-cofactor (e.g., a heme-containing protein) and one or more flavor precursor molecules include animal-based or non-animal (e.g., plant-based), or combinations of animal-based and non-animal-based, food products in the form of hot dogs, burgers, ground meat, sausages, steaks, filets, roasts, breasts, thighs, wings, meatballs, meatloaf, bacon, strips, fingers, nuggets, cutlets, or cubes.

Consumable food products for use in the present disclosure can be flavor additive compositions, e.g., for addition to another consumable food product before, during, or

after its cooking process. A flavor additive composition can include an iron complex such as a heme-cofactor (e.g., a heme-containing protein), and one or more flavor precursors.

A flavor additive composition can include a heme protein, e.g., an isolated and purified heme protein; such a flavor additive composition can be used to modulate the flavor and/or aroma profile of a consumable food product that comprises one or more flavor precursor molecules or compositions. A flavor additive composition can include one or more flavor precursor molecules or compositions; such a flavor additive composition can be used to modulate the flavor and/or aroma profile of a consumable food product that comprises the heme protein, e.g., an isolated and purified heme protein.

A flavor additive composition can be in the form, of but not limited to, soup or stew bases, bouillon, e.g., powder or cubes, flavor packets, or seasoning packets or shakers. Such flavor additive compositions can be used to modulate the flavor and/or aroma profile for a variety of consumable food products, and can be added to a consumable food product before, during, or after cooking of the consumable food product.

In some embodiments, a flavor additive composition such as one including an iron complex (e.g., ferrous chlorin or a heme protein) and one or more flavor precursors can be reacted (e.g., *in vitro*) with heating to generate a particular flavor and/or aroma profile of interest and the resultant product mixture can be added to the consumable food product of interest, which can then be eaten as-is or can be additionally modified, e.g., by additional cooking. In some embodiments, the iron complex can be removed from the resultant product mixture before adding the product mixture to the consumable food product of interest. For example, the iron complex can be removed from the product mixture using chromatographic techniques such as column chromatography, e.g., a column containing heme or iron-chlorin.

In some embodiments, the iron complex such as a heme-cofactor, e.g., a heme-protein, and the one or more flavor precursor flavor additive compositions can be soy-free, wheat-free, yeast-free, MSG-free, and free of protein hydrolysis products, and can taste meaty, highly savory, and without off odors or flavors.

In one aspect, this document features a food product that includes an iron complex such as a heme moiety, or a porphyrin, a porphyrinogen, a corrin, a corrinoid, a chlorin, a bacteriochorophyll, a corphin, a chlorophyllin, a bacteriochlorin, or an isobacteriochlorin moiety complexed to an iron ion and one or more flavor precursor molecules selected from the group consisting of glucose, fructose, ribose, arabinose, glucose-6-phosphate, fructose 6-phosphate, fructose 1,6-diphosphate, inositol, maltose, sucrose, maltodextrin, glycogen, nucleotide-bound sugars, molasses, a phospholipid, a lecithin, inosine, inosine monophosphate (IMP), guanosine monophosphate (GMP), pyrazine, adenosine monophosphate (AMP), lactic acid, succinic acid, glycolic acid, thiamine, creatine, pyrophosphate, vegetable oil, algal oil, corn oil, soybean oil, palm fruit oil, palm kernel oil, safflower oil, flaxseed oil, rice bran oil, cottonseed oil, sunflower oil, canola oil, olive oil, a free fatty acid, cysteine, methionine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine, proline, serine, tyrosine, glutathione, an amino acid derivative, a protein hydrolysate, a malt extract, a yeast extract, and a peptone. The heme moiety can be a heme-containing protein, a heme moiety bound to a non-peptidic polymer; or a heme moiety bound to a solid support. The heme-containing protein can be a plant, mammalian, a yeast or filamentous fungi, or bacterial heme-containing protein. The food product can include two to one hundred, two to fifty flavor precursors, two to forty flavor precursors, two to thirty-five flavor precursors, two to ten flavor precursors, or two to six flavor precursors. In some embodiments, the one or more flavor precursor molecules are selected from the group consisting of glucose, ribose, cysteine, a cysteine derivative, thiamine, alanine, methionine, lysine, a lysine derivative, glutamic acid, a glutamic acid derivative, IMP, GMP, lactic acid, maltodextrin, creatine, alanine, arginine, asparagine, aspartate, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, methionine, phenylalanine, proline, threonine, tryptophan, tyrosine, valine, linoleic acid, and mixtures thereof. The heme-containing protein can be a non-symbiotic hemoglobin or a leghemoglobin (e.g., a plant leghemoglobin such as one from soybean, alfalfa, lupin, pea, cow pea, or lupin). The heme-containing protein can include an amino acid sequence having at least 80% sequence identity to a polypeptide set forth in SEQ ID NOs:1-26.

The heme-containing protein can be isolated and purified. The food product further can include a food-grade oil, a seasoning agent, a flavoring agent, a protein, a protein concentrate, an emulsifier, a gelling agent, or a fiber. The food product can be a meat substitute, a soup base, stew base, snack food, bouillon powder, bouillon cube, a flavor packet, or a frozen food product. Any of the food products can be free of animal products. The food product can be sealed within a packet or shaker.

This document also features a method for producing a flavor compound. The method can include combining an iron complex (e.g., a heme moiety, a porphyrin, a porphyrinogen, a corrin, a corrinoid, a chlorin, a bacteriochorophyll, a corphin, a chlorophyllin, a bacteriochlorin, or an isobacteriochlorin complexed to an iron) and one or more flavor precursor molecules to form a mixture, the one or more flavor precursor molecules selected from the group consisting of glucose, fructose, arabinose, ribose glucose-6-phosphate, fructose 6-phosphate, fructose 1,6-diphosphate, inositol, maltose, sucrose, maltodextrin, glycogen, nucleotide-bound sugars, molasses, a phospholipid, a lecithin, inosine, inosine monophosphate (IMP), guanosine monophosphate (GMP), pyrazine, adenosine monophosphate (AMP), lactic acid, succinic acid, glycolic acid, thiamine, creatine, pyrophosphate, vegetable oil, algal oil, corn oil, soybean oil, palm fruit oil, palm kernel oil, safflower oil, flaxseed oil, rice bran oil, cottonseed oil, canola oil, olive oil, sunflower oil, flaxseed oil, coconut oil, mango oil, a free fatty acid, cysteine, methionine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine, proline, serine, tyrosine, glutathione, an amino acid derivative, a protein hydrolysate, a malt extract, a yeast extract, and a peptone; and heating the mixture to form one or more flavor compounds selected from the group consisting of phenylacetaldehyde, 1-octen-3-one, 2-n-heptylfuran, 2-thiophenecarboxaldehyde, 3-thiophenecarboxaldehyde, butyrolactone, 2-undecenal, pyrazine, methyl-, furfural, 2-decanone, pyrrole, 1-octen-3-ol, 2-acetylthiazole, (E)-2-octenal, decanal, benzaldehyde, (E)-2-nonenal, pyrazine, 1-hexanol, 1-heptanol, dimethyl trisulfide, 2-nonanone, 2-pentanone, 2-heptanone, 2,3-butanedione, heptanal, nonanal, 2-octanone, 1-octanol, 3-ethylcyclopentanone, 3-octen-2-one, (E,E)-2,4-heptadienal, (Z)-2-heptenal, 2-heptanone, 6-methyl-, (Z)-4-heptenal,

(E,Z)-2,6-nonadienal, 3-methyl-2-butenal, 2-pentyl-furan, thiazole, (E, E)-2,4-decadienal,, hexanoic acid, 1-ethyl-5-methylcyclopentene, (E,E)-2,4-nonadienal, (Z)-2-decenal, dihydro-5-pentyl-2(3H)-furanone, trans-3-nonen-2-one, (E,E)-3,5-octadien-2-one, (Z)-2-octen-1-ol, 5-ethyldihydro-2(3H)-furanone, 2-butenal, 1-penten-3-ol, (E)-2-hexenal, formic acid, heptyl ester, 2-pentyl-thiophene, (Z)-2-nonenal, 2-hexyl-thiophene, (E)-2-decenal, 2-ethyl-5-methyl-pyrazine, 3-ethyl-2,5-dimethyl-pyrazine, 2-ethyl-1-hexanol, thiophene, 2-methyl-furan, pyridine, butanal, 2-ethyl-furan, 3-methyl-butanal, trichloromethane, 2-methyl-butanal, methacrolein, 2-methyl-propanal, propanal, acetaldehyde, 2-propyl-furan, dihydro-5-propyl-2(3H)- furanone, 1,3-hexadiene, 4-decyne, pentanal, 1-propanol, heptanoic acid, trimethyl-ethanethiol, , 1-butanol, 1-penten-3-one, dimethyl sulfide, 2-ethyl furan, 2-pentyl-thiophene, 2-propenal, 2-tridecen-1-ol, 4-octene, 2-methyl thiazole,, methyl-pyrazine, 2-butanone, 2-pentyl-furan, 2-methyl-propanal, butyrolactone,, 3-methyl-butanal, methyl-thiirane, 2-hexyl-furan, butanal,, 2-methyl-butanal, 2-methyl-furan, furan, octanal, 2-heptenal, 1-octene, formic acid heptyl ester, 3-pentyl-furan, and 4-penten-2-one. The heme moiety can be a heme-containing protein, a heme moiety bound to a non-peptidic polymer; or a heme moiety bound to a solid support. The method can include combining cysteine, ribose, lactic acid, lysine, and/or thiamine with the heme-containing protein.

In another aspect, this document features a method for producing a flavor compound. The method includes combining an iron complex, such as a heme-containing protein, and one or more flavor precursor molecules to form a mixture, the one or more flavor precursor molecules selected from the group consisting of glucose, fructose, ribose, arabinose, glucose-6-phosphate, fructose 6-phosphate, fructose 1,6-diphosphate, inositol, maltose, sucrose, maltodextrin, glycogen, nucleotide-bound sugars, molasses, a phospholipid, a lecithin, inosine, IMP, GMP, pyrazine, AMP, lactic acid, succinic acid, glycolic acid, thiamine, creatine, pyrophosphate, vegetable oil, algal oil, corn oil, soybean oil, palm fruit oil, palm kernel oil, safflower oil, flaxseed oil, rice bran oil, cottonseed oil, olive oil, sunflower oil, canola oil, flaxseed oil, coconut oil, mango oil, a free fatty acid, methionine, cysteine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine,

proline, serine, tyrosine, glutathione, an amino acid derivative, a protein hydrolysate, a malt extract, a yeast extract, and a peptone; and heating the mixture to form one or more flavor compounds set forth in Tables 3, 8, or 9. For example, the flavor precursors can include cysteine, a sugar, and one or more other precursors.

This document also features a method for imparting a meat like flavor (e.g., beef-like, chicken like, pork-like, lamb-like, turkey-like, duck-like, deer-like, or bison-like) to a food product. The method includes contacting the food product with a flavoring composition, the flavoring composition comprising i) an iron complex, such as a heme moiety (e.g., a heme-containing protein); and ii) one or more flavor precursor molecules selected from the group consisting of glucose, fructose, ribose, arabinose, glucose-6-phosphate, fructose 6-phosphate, fructose 1,6-diphosphate, inositol, maltose, sucrose, maltodextrin, glycogen, nucleotide-bound sugars, molasses, a phospholipid, a lecithin, inosine, IMP, GMP, pyrazine, AMP, lactic acid, succinic acid, glycolic acid, thiamine, creatine, pyrophosphate, vegetable oil, algal oil, corn oil, soybean oil, palm fruit oil, palm kernel oil, safflower oil, flaxseed oil, rice bran oil, cottonseed oil, olive oil, sunflower oil, canola oil, flaxseed oil, coconut oil, mango oil, a free fatty acid, cysteine, methionine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine, proline, serine, tyrosine, glutathione, an amino acid derivative, a protein hydrolysate, a malt extract, a yeast extract, and a peptone; wherein after heating the food product and the flavoring composition together, a meat like flavor (e.g., beef-like, chicken like, pork-like, lamb-like, turkey-like, duck-like, deer-like, or bison-like) is imparted to the food product. In some embodiments, the iron complex is removed from the food product. The flavoring composition further can include a seasoning agent, a flavoring agent, a protein, a protein concentrate, or an emulsifier. The flavoring composition can be sealed within a packet or shaker.

In another aspect, this document features a method of making a food product. The method includes combining an isolated heme-containing protein and one or more flavor precursor molecules to form a mixture, the one or more flavor precursor molecules selected from the group consisting of glucose, fructose, ribose, arabinose, glucose-6-

phosphate, fructose 6-phosphate, fructose 1,6-diphosphate, inositol, maltose, sucrose, maltodextrin, glycogen, nucleotide-bound sugars, molasses, a phospholipid, a lecithin, inosine, IMP, GMP, pyrazine, AMP, lactic acid, succinic acid, glycolic acid, thiamine, creatine, pyrophosphate, sunflower oil, coconut oil, canola oil, flaxseed oil, mango oil, a free fatty acid, cysteine, methionine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine, proline, serine, tyrosine, glutathione, an amino acid derivative, a protein hydrolysate, a malt extract, a yeast extract, and a peptone; and heating the mixture.

Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention pertains. Although methods and materials similar or equivalent to those described herein can be used to practice the invention, suitable methods and materials are described below. All publications, patent applications, patents, and other references mentioned herein are incorporated by reference in their entirety. In case of conflict, the present specification, including definitions, will control. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

The details of one or more embodiments of the invention are set forth in the accompanying drawings and the description below. Other features, objects, and advantages of the invention will be apparent from the description and drawings, and from the claims. The word "comprising" in the claims may be replaced by "consisting essentially of" or with "consisting of," according to standard practice in patent law.

DESCRIPTION OF THE DRAWINGS

FIG. 1 contains amino acid sequences of exemplary heme-containing proteins.

FIG. 2 is a bar graph of the beefiness rating of the meat replica with or without the Magic Mix, both samples in triplicate with 1% w/v LegH protein. Tasters rated beefiness on a scale from 1-7, with 1 being not beefy at all and 7 being exactly like ground beef.

DETAILED DESCRIPTION

This document is based on methods and materials for modulating the taste and/or aroma profile of food products. As described herein, compositions containing one or more flavor precursors and one or more highly conjugated heterocyclic rings complexed to an iron (referred to herein as an iron complex) can be used to modulate the taste and/or aroma profile of food products. Such iron complexes include heme moieties or other highly conjugated heterocylic rings complexed to an iron ion (referred to as an iron complex). "Heme" refers to a prosthetic group bound to iron (Fe^{2+} or Fe^{3+}) in the center of a porphyrin ring. Thus, an iron complex can be a heme moiety, or a porphyrin, porphyrinogen, corrin, corrinoid, chlorin, bacteriochorophyll, corphin, chlorophyllin, bacteriochlorin, or isobacteriochlorin moiety complexed to iron ion. The heme moiety that can be used to modulate the taste and/or aroma profile of food products can be a heme cofactor such as a heme-containing protein; a heme moiety bound to a non-peptidic polymer or other macromolecule such as a liposome, a polyethylene glycol, a carbohydrate, a polysaccharide, a cyclodextrin, a polyethylenimine, a polyacrylate, or derivatives thereof; a siderophore (i.e., an iron chelating compound); or a heme moiety bound to a solid support (e.g., beads) composed of a chromatography resin, cellulose, graphite, charcoal, or diatomaceous earth.

In some embodiments, the iron complexes catalyze some reactions and produce flavor precursors without heating or cooking. In some embodiments, the iron complex destabilizes upon heating or cooking and releases the iron, e.g., the protein is denatured, so flavor precursors can be generated.

Suitable flavor precursors include sugars, sugar alcohols, sugar derivatives, oils (e.g., vegetable oils), free fatty acids, alpha-hydroxy acids, dicarboxylic acids, amino acids and derivatives thereof, nucleosides, nucleotides, vitamins, peptides, protein hydrolysates, extracts, phospholipids, lecithin, and organic molecules. Non-limiting examples of such flavor precursors are provided in Table 1.

TABLE 1

Flavor Precursor Molecules

Sugars, sugar alcohols, sugar acids, and sugar derivatives: glucose, fructose, ribose, sucrose, arabinose, glucose-6-phosphate,fructose-6-phosphate, fructose 1,6-diphosphate, inositol, maltose, molasses, maltodextrin, glycogen, galactose, lactose, ribitol, gluconic acid and glucuronic acid, amylose, amylopectin, or xylose
Oils: coconut oil, mango oil, sunflower oil, cottonseed oil, safflower oil, rice bran oil, cocoa butter, palm fruit oil, palm oil, soybean oil, canola oil, corn oil, sesame oil, walnut oil, flaxseed, jojoba oil, castor, grapeseed oil, peanut oil, olive oil, algal oil, oil from bacteria or fungi
Free fatty acids: caprylic acid, capric acid, lauric acid, myristic acid, palmititic acid, palmitoleic acid, stearic, oleic acid, linoleic acid, alpha linolenic acid, gamma linolenic acid, arachidic acid, arachidonic acid, behenic acid, or erucic acid
Amino acids and derivatives thereof: cysteine, cystine, a cysteine sulfoxide, allicin, selenocysteine, methionine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, 5-hydroxytryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine, proline, serine, or tyrosine
Nucleosides and Nucleotides: inosine , inosine monophosphate (IMP), guanosine, guanoside monophosphate (GMP), adenosine, adenosine monophosophate (AMP)
Vitamins: thiamine, vitamin C, Vitamin D, Vitamin B6, or Vitamin E
Misc: phospholipid, lecithin, pyrazine, creatine, pyrophosphate
Acids: acetic acid, alpha hydroxy acids such as lactic acid or glycolic acid, tricarboxylic acids such as citric acid, dicarboxylic acids such as succinic acid or tartaric acid
Peptides and protein hydrolysates: glutathione, vegetable protein hydrolysates, soy protein hydrolysates, yeast protein hydrolysates, algal protein hydrolysatess,

meat protein hydrolysates
Extracts: a malt extract, a yeast extract, and a peptone

In some embodiments, one flavor precursor or combinations of two to one hundred flavor precursors, two to ninety, two to eighty, two to seventy, two to sixty, or two to fifty flavor precursors are used. For example, combinations of two to forty flavor precursors, two to thirty-five flavor precursors, two to ten flavor precursors, or two to six flavor precursors can be used with the one or more iron complexes (e.g., heme co-factors such as a heme-containing proteins). For example, the one or more flavor precursors can be glucose, ribose, cysteine, a cysteine derivative, thiamine, lysine, a lysine derivative, glutamic acid, a glutamic acid derivative, alanine, methionine, IMP, GMP, lactic acid, and mixtures thereof (e.g., glucose and cysteine; cysteine and ribose; cysteine, glucose or ribose, and thiamine; cysteine, glucose or ribose, IMP, and GMP; cysteine, glucose or ribose, and lactic acid). For example, the one or more flavor precursors can be alanine, arginine, asparagine, aspartate, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, threonine, tryptophan, tyrosine, valine, glucose, ribose, maltodextrin, thiamine, IMP, GMP, lactic acid, and creatine.

As used herein, the term “heme containing protein” can be used interchangeably with “heme containing polypeptide” or “heme protein” or “heme polypeptide” and includes any polypeptide that can covalently or noncovalently bind a heme moiety. In some embodiments, the heme-containing polypeptide is a globin and can include a globin fold, which comprises a series of seven to nine alpha helices. Globin type proteins can be of any class (e.g., class I, class II, or class III), and in some embodiments, can transport or store oxygen. For example, a heme-containing protein can be a non-symbiotic type of hemoglobin or a leghemoglobin. A heme-containing polypeptide can be a monomer, i.e., a single polypeptide chain, or can be a dimer, a trimer, tetramer, and/or higher order oligomers. The life-time of the oxygenated $Fe2^{+}$ state of a heme-containing protein can be similar to that of myoglobin or can exceed it by 10%, 20%, 30% 50%, 100% or more under conditions in which the heme-protein-containing consumable is manufactured,

stored, handled or prepared for consumption. The life-time of the unoxygenated Fe^{2+} state of a heme-containing protein can be similar to that of myoglobin or can exceed it by 10%, 20%, 30% 50%, 100% or more under conditions in which the heme-protein-containing consumable is manufactured, stored, handled or prepared for consumption

Non-limiting examples of heme-containing polypeptides can include an androglobin, a cytoglobin, a globin E, a globin X, a globin Y, a hemoglobin, a myoglobin, an erythrocruorin, a beta hemoglobin, an alpha hemoglobin, a protoglobin, a cyanoglobin, a cytoglobin, a histoglobin, a neuroglobins, a chlorocruorin, a truncated hemoglobin (e.g., HbN or HbO), a truncated 2/2 globin, a hemoglobin 3 (e.g., Glb3), a cytochrome, or a peroxidase.

Heme-containing proteins that can be used in the compositions and food products described herein can be from mammals (e.g., farms animals such as cows, goats, sheep, pigs, ox, or rabbits), birds, plants, algae, fungi (e.g., yeast or filamentous fungi), ciliates, or bacteria. For example, a heme-containing protein can be from a mammal such as a farm animal (e.g., a cow, goat, sheep, pig, ox, or rabbit) or a bird such as a turkey or chicken. Heme-containing proteins can be from a plant such as *Nicotiana tabacum* or *Nicotiana sylvestris* (tobacco); *Zea mays* (corn), *Arabidopsis thaliana*, a legume such as *Glycine max* (soybean), *Cicer arietinum* (garbanzo or chick pea), *Pisum sativum* (pea) varieties such as garden peas or sugar snap peas, *Phaseolus vulgaris* varieties of common beans such as green beans, black beans, navy beans, northern beans, or pinto beans, *Vigna unguiculata* varieties (cow peas), *Vigna radiata* (Mung beans), *Lupinus albus* (lupin), or *Medicago sativa* (alfalfa); *Brassica napus* (canola); *Triticum sps.* (wheat, including wheat berries, and spelt); *Gossypium hirsutum* (cotton); *Oryza sativa* (rice); *Zizania sps.* (wild rice); *Helianthus annuus* (sunflower); *Beta vulgaris* (sugarbeet); *Pennisetum glaucum* (pearl millet); *Chenopodium sp.* (quinoa); *Sesamum sp.* (sesame); *Linum usitatissimum* (flax); or *Hordeum vulgare* (barley). Heme-containing proteins can be isolated from fungi such as *Saccharomyces cerevisiae*, *Pichia pastoris*, *Magnaporthe oryzae*, *Fusarium graminearum*, *Aspergillus oryzae*, *Trichoderma reesei*, *Myceliopthera thermophile*, *Kluyvera lactis*, or *Fusarium oxysporum*. Heme-containing proteins can be isolated from bacteria such as *Escherichia coli*, *Bacillus subtilis, Bacillus licheniformis,*

Bacillus megaterium, Synechocistis sp., Aquifex aeolicus, Methylacidiphilum infernorum, or thermophilic bacteria such as *Thermophilus*. The sequences and structure of numerous heme-containing proteins are known. See for example, Reedy, *et al.*, *Nucleic Acids Research*, 2008, Vol. 36, Database issue D307–D313 and the Heme Protein Database available on the World Wide Web at hemeprotein.info/heme.php.

For example, a non-symbiotic hemoglobin can be from a plant selected from the group consisting of soybean, sprouted soybean, alfalfa, golden flax, black bean, black eyed pea, northern, garbanzo, moong bean, cowpeas, pinto beans, pod peas, quinoa, sesame, sunflower, wheat berries, spelt, barley, wild rice, or rice.

Any of the heme-containing proteins described herein that can be used for producing food products can have at least 70% (e.g., at least 75%, 80%, 85%, 90%, 95%, 97%, 98%, 99%, or 100%) sequence identity to the amino acid sequence of the corresponding wild-type heme-containing protein or fragments thereof that contain a heme-binding motif. For example, a heme-containing protein can have at least 70% sequence identity to an amino acid sequence set forth in FIG. 1, including a non-symbiotic hemoglobin such as that from *Vigna radiata* (SEQ ID NO:1), *Hordeum vulgare* (SEQ ID NO:5), *Zea mays* (SEQ ID NO:13), *Oryza sativa subsp. japonica (rice)* (SEQ ID NO:14), or *Arabidopsis thaliana* (SEQ ID NO:15), a Hell's gate globin I such as that from *Methylacidiphilum infernorum* (SEQ ID NO:2), a flavohemoprotein such as that from *Aquifex aeolicus* (SEQ ID NO:3), a leghemoglobin such as that from *Glycine max* (SEQ ID NO:4), *Pisum sativum* (SEQ ID NO:16), or *Vigna unguiculata* (SEQ ID NO:17), a heme-dependent peroxidase such as from *Magnaporthe oryzae*, (SEQ ID NO:6) or *Fusarium oxysporum* (SEQ ID NO:7), a cytochrome c peroxidase from *Fusarium graminearum* (SEQ ID NO:8), a truncated hemoglobin from *Chlamydomonas moewusii* (SEQ ID NO:9), *Tetrahymena pyriformis* (SEQ ID NO:10, group I truncated), *Paramecium caudatum* (SEQ ID NO:11, group I truncated), a hemoglobin from *Aspergillus niger* (SEQ ID NO:12), or a mammalian myoglobin protein such as the *Bos taurus* (SEQ ID NO:18) myoglobin, *Sus scrofa* (SEQ ID NO:19) myoglobin, *Equus caballus* (SEQ ID NO:20) myoglobin, a heme-protein from *Nicotiana benthamiana* (SEQ ID NO:21), *Bacillus subtilis* (SEQ ID NO:22), *Corynebacterium glutamicum* (SEQ ID

NO:23), Synechocystis PCC6803 (SEQ ID NO:24), Synechococcus sp. PCC 7335 (SEQ ID NO:25), or *Nostoc commune* (SEQ ID NO:26).

The percent identity between two amino acid sequences can be determined as follows. First, the amino acid sequences are aligned using the BLAST 2 Sequences (Bl2seq) program from the stand-alone version of BLASTZ containing BLASTP version 2.0.14. This stand-alone version of BLASTZ can be obtained from Fish & Richardson's web site (e.g., www.fr.com/blast/) or the U.S. government's National Center for Biotechnology Information web site (www.ncbi.nlm.nih.gov). Instructions explaining how to use the Bl2seq program can be found in the readme file accompanying BLASTZ. Bl2seq performs a comparison between two amino acid sequences using the BLASTP algorithm. To compare two amino acid sequences, the options of Bl2seq are set as follows: -i is set to a file containing the first amino acid sequence to be compared (e.g., C:\seq1.txt); -j is set to a file containing the second amino acid sequence to be compared (e.g., C:\seq2.txt); -p is set to blastp; -o is set to any desired file name (e.g., C:\output.txt); and all other options are left at their default setting. For example, the following command can be used to generate an output file containing a comparison between two amino acid sequences: C:\Bl2seq –i c:\seq1.txt –j c:\seq2.txt –p blastp –o c:\output.txt. If the two compared sequences share homology, then the designated output file will present those regions of homology as aligned sequences. If the two compared sequences do not share homology, then the designated output file will not present aligned sequences. Similar procedures can be following for nucleic acid sequences except that blastn is used.

Once aligned, the number of matches is determined by counting the number of positions where an identical amino acid residue is presented in both sequences. The percent identity is determined by dividing the number of matches by the length of the full-length polypeptide amino acid sequence followed by multiplying the resulting value by 100. It is noted that the percent identity value is rounded to the nearest tenth. For example, 78.11, 78.12, 78.13, and 78.14 is rounded down to 78.1, while 78.15, 78.16, 78.17, 78.18, and 78.19 is rounded up to 78.2. It also is noted that the length value will always be an integer.

It will be appreciated that a number of nucleic acids can encode a polypeptide having a particular amino acid sequence. The degeneracy of the genetic code is well known to the art; i.e., for many amino acids, there is more than one nucleotide triplet that serves as the codon for the amino acid. For example, codons in the coding sequence for a given enzyme can be modified such that optimal expression in a particular species (e.g., bacteria or fungus) is obtained, using appropriate codon bias tables for that species.

Heme-containing proteins can be extracted from the source material (e.g., extracted from animal tissue, or plant, fungal, algal, or bacterial biomass, or from the culture supernatant for secreted proteins) or from a combination of source materials (e.g., multiple plant species). Leghemoglobin is readily available as an unused by-product of commodity legume crops (e.g., soybean, alfalfa, or pea). The amount of leghemoglobin in the roots of these crops in the United States exceeds the myoglobin content of all the red meat consumed in the United States.

In some embodiments, extracts of heme-containing proteins include one or more non-heme-containing proteins from the source material (e.g., other animal, plant, fungal, algal, or bacterial proteins) or from a combination of source materials (e.g., different animal, plant, fungi, algae, or bacteria).

In some embodiments, heme-containing proteins are isolated and purified from other components of the source material (e.g., other animal, plant, fungal, algal, or bacterial proteins). As used herein, the term "isolated and purified" indicates that the preparation of heme-containing protein is at least 60% pure, e.g., greater than 65%, 70%, 75%, 80%, 85%, 90%, 95%, or 99% pure. Without being bound by theory, isolating and purifying proteins can allow the food products to be made with greater consistency and greater control over the properties of the food product as unwanted material is eliminated. Proteins can be separated on the basis of their molecular weight, for example, by size exclusion chromatography, ultrafiltration through membranes, or density centrifugation. In some embodiments, the proteins can be separated based on their surface charge, for example, by isoelectric precipitation, anion exchange chromatography, or cation exchange chromatography. Proteins also can be separated on the basis of their solubility, for example, by ammonium sulfate precipitation, isoelectric precipitation, surfactants,

detergents or solvent extraction. Proteins also can be separated by their affinity to another molecule, using, for example, hydrophobic interaction chromatography, reactive dyes, or hydroxyapatite. Affinity chromatography also can include using antibodies having specific binding affinity for the heme-containing protein, nickel NTA for His-tagged recombinant proteins, lectins to bind to sugar moieties on a glycoprotein, or other molecules which specifically binds the protein.

Heme-containing proteins also can be recombinantly produced using polypeptide expression techniques (e.g., heterologous expression techniques using bacterial cells, insect cells, fungal cells such as yeast, plant cells such as tobacco, soybean, or *Arabidopsis*, or mammalian cells). In some cases, standard polypeptide synthesis techniques (e.g., liquid-phase polypeptide synthesis techniques or solid-phase polypeptide synthesis techniques) can be used to produce heme-containing proteins synthetically. In some cases, *in vitro* transcription-translation techniques can be used to produce heme-containing proteins.

The protein used in the consumable may be soluble in a solution. In some embodiments, the isolated and purified proteins are soluble in solution at greater than 5, 10, 15, 20, 25, 50, 100, 150, 200, or 250g/L.

In some embodiments, the isolated and purified protein is substantially in its native fold and water soluble. In some embodiments, the isolated and purified protein is more than 50, 60, 70, 80, or 90% in its native fold. In some embodiments, the isolated and purified protein is more than 50, 60, 70, 80, or 90% water soluble.

In some embodiments, the food product contains between 0.01% and 5% by weight of a heme protein. In some embodiments, the food product contains between 0.01% and 5% by weight of leghemoglobin. Some meat also contains myoglobin, a heme protein, which accounts for most of the red color and iron content of some meat. It 20 is understood that these percentages can vary in meat and the food products can be produced to approximate the natural variation in meat.

In some embodiments, the food product comprises about 0.05%, about 0.1%, about 0.2%, about 0.3%, about 0.4%, about 0.5%, about 0.6%, about 0.7%, about 0.8%, about 0.9%, about 5 1%, about 1.1%, about 1.2%, about 1.3%, about 1.4%, about 1.5%,

about 1.6%, about 1.7%, about 1.8%, about 1.9%, about 2%, or more than about 2% of an iron-carrying protein (e.g., a heme-containing protein) by dry weight or total weight. In some cases, the iron carrying protein has been isolated and purified from a source.

Modulating Flavor and/or Aroma Profiles

As described herein, different combinations of flavor precursors can be used with one or more iron complexes (e.g., a ferrous chlorin, a chlorin-iron complex, or a heme-cofactor such as a heme-containing protein or heme bound to a non-peptidic polymer such as polyethylene glycol or to a solid support) to produce different flavor and aroma profiles when the flavor precursors and iron complexes are heated together (e.g., during cooking). The resultant flavor and/or aroma profile can be modulated by the type and concentration of the flavor precursors, the pH of the reaction, the length of cooking, the type and amount of iron complex (e.g., a heme-cofactor such as heme-containing protein, heme bound to non-peptidic polymer or macromolecule, or heme bound to a solid support), the temperature of the reaction, and the amount of water activity in the product, among other factors. In embodiments in which a heme moiety is bound to a solid support such as cellulose or a chromatography resin, graphite, charcoal, or diatomaceous earth, the solid support (e.g., beads) can be incubated with sugars and/or one or more other flavor precursors to generate flavors, and then the solid support with attached heme moiety can be re-used, i.e., incubated again with sugars and/or one or more other flavor precursors to generate flavors.

Table 2 provides non-limiting examples of flavor types that can be generated by combining one or more flavor precursors and one or more heme co-factors (e.g., heme-containing proteins). See also Tables 7 and/or 11.

TABLE 2

Flavor Types	
beef	beef broth
beef dripping	cheesy

cold-cut deli meat	squash
bacon	sharp
meaty	fruity
brothy	floral
ramen	musty
egg	fried food
malty	caramel
bready	barbeque
sulfur	chocolate
fried chicken	sweet
browned	potato
pretzel	french toast
grassy	breadcrust
bloody	mushroom
broccoli	chicken
brothy	cumin
buttery	umami
metallic	raisin
yeasty	goaty
vegetable broth	

Flavor and aroma profiles are created by different chemical compounds formed by chemical reactions between the heme co-factor (e.g., heme-containing protein) and flavor precursors. Gas chromatography–mass spectrometry (GCMS) can be used to separate and identify the different chemical compounds within a test sample. For example, volatile chemicals can be isolated from the head space after heating a heme-containing protein and one or more flavor precursors.

Table 3 provides non-limiting examples of compounds that can be produced. See also Tables 8, 9, 12, and/or 14.

TABLE 3

Compounds Produced

phenylacetaldehyde	2-butenal, 2-ethyl-	1,3-hexadiene
1-octen-3-one	acetonitrile	4-decyne
2-n-heptylfuran		pentanal
2-thiophenecarboxaldehyde	(E)-2-Hexenal	1-propanol
3-thiophenecarboxaldehyde	4-ethyl-phenol,	heptanoic acid
1-octene	3-octanone	ethanethiol
butyrolactone	styrene	2-methyl-1-heptene
2-undecenal	furan, 3-pentyl-	(E)-4-octene
propyl-cyclopropane	formic acid, heptyl ester	2-methyl-2-heptene
methyl-pyrazine	(E)-2-Heptenal	pentanoic acid
1-hydroxy-propanone	6-methyl-5-hepten-2-one	nonanoic acid
acetic acid	n-caproic acid vinyl ester	1,3-dimethyl-benzene
furfural	2-ethyl-2-hexenal	
2-decanone	1-hepten-3-ol	toluene
pyrrole	1-ethyl-1-methyl-cyclopentane	1-butanol
1-octen-3-ol	3-ethyl-2-methyl-1,3-hexadiene	2,3,3-trimethyl-pentane
2-acetylthiazole	2-pentyl-thiophene	isopropyl alcohol
(E)-2-octenal	(Z)-2-nonenal	2,2,4,6,6-pentamethyl-heptane
decanal	2-n-octylfuran	phenol
benzaldehyde	2-hexyl-thiophene	1-penten-3-one
(E)-2-Nonenal	4-cyclopentene-1,3-dione	dimethyl sulfide
pyrazine	1-nonanol	thiirane
1-pentanol	(E)-2-decenal	(E)-2-octen-1-ol
trans-2-(2-pentenyl)furan	4-ethyl-benzaldehyde	2,4-dimethyl-1-heptene
1-hexanol	1,7-octadien-3-ol	1,3-bis(1,1-dimethylethyl)-benzene
1-heptanol	octanoic acid	heptane
dimethyl trisulfide	2-ethyl-5-methyl-pyrazine	4,7-dimethyl-undecane
2-nonanone	3-ethyl-2,5-dimethyl-pyrazine	acetophenone
2-pentanone	1,3,5-cycloheptatriene	tridecane
2-heptanone	2-ethyl-1-hexanol	thiophosphoramide, s-methyl ester
2,3-butanedione	4-methyl-octanoic acid	2-methyl-thiazole

heptanal	m-aminophenylacetylene	3-(1-methylethoxy)-propanenitrile,
nonanal	benzene	2,4-bis(1,1-dimethylethyl)-phenol
2-octanone	thiophene	3-ethyl-2,2-dimethyl-pentane
2-butanone	2-methyl-furan	3-ethyl-pentane
octanal	pyridine	2,3,4-trimethyl-pentane
1-octanol	furan	2,4,6-trimethyl-octane
3-ethylcyclopentanone	butanal	2,6-dimethyl-nonane
8-methyl-1-undecene	2-ethyl-furan	2-hexyl-furan
3-octen-2-one	carbon disulfide	4-methyl-5-thiazoleethanol
2,4-Heptadienal, (E,E)-	Furan, 2-hexyl-:2	4-penten-2-one
(Z)-2-heptenal	3-methyl-butanal	4-methylthiazole
6-methyl-2-heptanone	2-methyl-butanal	2-methyl-3-pentanone
(Z)-4-heptenal	methacrolein	2,3-pentanedione
(E,Z)-2,6-nonadienal	octane	(E)-2-tridecen-1-ol
3-methyl-2-butenal	ethanol	2-thiophenemethanamine
2-pentyl-furan	2-methyl-propanal	(Z)-2-nonenal,
thiazole	acetone	methyl thiolacetate
(E,E)-2,4-decadienal	propanal	methyl ethanoate
hexanoic acid	methyl-thiirane	isothiazole
1-ethyl-5-methylcyclopentene	acetaldehyde	3,3-dimethyl-hexane
(E,E)-2,4-nonadienal	2-propenal	4-methyl-heptane
(Z)-2-decenal	2-propyl-furan	2,4-dimethyl-heptane
dihydro-5-pentyl-2(3h)-furanone	dihydro-5-propyl-2(3H)-furanone	2,3,4-trimethyl-heptane
trans-3-nonen-2-one	dihydro-3-(2H)-thiophenone	2-methyl-heptane
(E,E)-3,5-octadien-2-one	2,2,6-trimethyl-decane	2-methyl-3-furanthiol
(Z)-2-octen-1-ol	3,3'-dithiobis[2-methyl-furan	4-amino-1,2,5-oxadiazole-3-carbonitrile
5-ethyldihydro-2(3h)-furanone	1-heptene	1,2-benzisothiazol-3(2H)-one
2-butenal	1,3-octadiene	2-acetyl-propen-2-ol,
1-penten-3-ol	1-nonene	1-decen-3-one
1-(ethylthio)-2-(methylthio)-buta-1,3-diene		

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein is heated in the presence of ground chicken, to increase specific volatile flavor and odorant components typically elevated in beef. For example, propanal, butanal, 2-ethyl-furan, heptanal, octanal, trans-2-(2-pentenyl)furan, (Z)-2-heptenal, (E)-2-octenal, pyrrole, 2,4-dodecadienal, 1-octanal, (Z)-2-decenal, or 2-undecenal can be increased in the presence of the heme-containing protein, which can impart a more beefy flavor to the chicken.

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein is heated in the presence of cysteine and glucose or other combinations of flavor precursors to provide a different profile of volatile odorants than when any subset of the three components are used individually. Volatile flavor components that are increased under these conditions include but are not limited to furan, acetone, thiazole, furfural, benzaldehyde, 2-pyridinecarboxaldehyde, 5-methyl-2-thiophenecarboxaldehyde, 3-methyl-2-thiophenecarboxaldehyde, 3-thiophenemethanol and decanol. See, e.g., Tables 8 and 9. Under these conditions, cysteine and glucose alone or in the presence of iron salts such as ferrous glucanate produced a sulfurous, odor, but addition of heme-containing proteins reduced the sulfurous odor and replaced it with flavors including but not limited to chicken broth, burnt mushroom, molasses, and bread.

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein is heated in the presence of cysteine and ribose to provide a different profile of volatile odorants. Heating in the presence of ribose created some additional compounds as compared to when a heme-containing protein and glucose were heated together. See Tables 8 and 9.

In some embodiments, an iron complex (e.g., a ferrous chlorophillin or a heme-cofactor such as a heme-containing protein) described herein can be heated in the presence of thiamine and a sugar to affect the formation of 5-Thiazoleethanol, 4-methyl-furan, 3,3'-dithiobis[2-methyl-furan, and/or 4-Methylthiazole. These compounds are known to be present in meat and have beefy, meaty taste notes.

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein can be heated in the presence of a nucleotide such as inosine monophosphate and/or guanosine monophosphate to control the formation of flavor compounds such as (E)-4-octene, 2-ethyl-furan, 2-pentanone, 2,3-butanedione, 2-methyl-thiazole, methyl-pyrazine, tridecane, (E)-2-octenal, 2-thiopenecarboxaldehyde, and/or 3-thiopenecarboxaldehyde. These compounds are known to be present in meat and have a beefy, meaty, buttery, and or savory flavor notes.

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein can be heated in the presence of lysine, a sugar such as ribose, and cysteine to control the formation of flavor compounds such as dimethyl trisulfide, nonanal, 2-pentyl thiophene, 2-nonenal furfural, 1-octanol, 2-nonenal, thiazole, 2-acetylthiazole, phenylacetaldehyde, and/or 2-acetylthiazole. These compounds are known to be present in meat and some have a beefy, meaty, and or savory flavor.

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein can be heated in the presence of lactic acid, a sugar such as ribose, and cysteine to control the formation of the flavor compounds nonanal, thiazole, 2-acetylthiazole, and/or 8-methyl 1-undecene. These compounds are known to be present in meat and have beefy, savory, browned, bready, and malty notes.

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein can be heated in the presence of amino acids, sugars such as glucose, ribose, and maltodextrin, lactic acid, thiamine, IMP, GMP, creatine, and salts such as potassium chloride and sodium chloride, to control the formation of flavor compounds such as 1,3-bis(1,1-dimethylethyl)-benzene, 2-methyl 3-furanthiol, and/or bis(2-methyl-4,5-dihydro-3-furyl) disulfide. These compounds are known to be present in meat and have beefy notes. See also Table 14.

In some embodiments, a particular type of heme-containing protein is chosen to control the formation of flavor compounds. See, for example, the results of Table 9, which shows that the addition of different types of heme-proteins (LegH, Barley, *B.*

myoglobin, or *A. aeolicus*) in flavor reaction mixtures containing one or more flavor precursor compounds results in many of the same key meat flavors, including but not limited to pentanone, 3-methyl butanal, 2-methyl butanal, 2-heptenal, 1-octene, nonanal, 2-propenal, 2-decenal, 2-nonanone, 2-octanone, 2-tridecen-1-ol, 2-octanone, 2-octenal, 4-methyl-2-heptanone, octanal, 2-undecenal, butyrolactone, 1-octen-3-one, 3-methylheptyl acetate, and 2-pentyl-thiophene. These differences in flavor compounds can change the overall taste profile.

In some embodiments, an iron complex (e.g., a ferrous chlorin or a heme-cofactor such as a heme-containing protein) described herein and one or more flavor precursors can be reacted (e.g., *in vitro*) with heating to generate a particular flavor and/or aroma profile of interest and the resultant flavor additive composition can be added to the consumable food product of interest, which can then be eaten as-is or can be additionally modified, e.g., by additional cooking.

In some embodiments, any undesirable flavors can be minimized by deodorizing with activated charcoal or by removing enzymes such as lipoxygenases (LOX), which can be present in trace amounts when using preparations of plant proteins, and which can convert unsaturated triacylglycerides (such as linoleic acid or linolenic acid) into smaller and more volatile molecules. LOX are naturally present in legumes such as peas, soybeans, and peanuts, as well as rice, potatoes, and olives. When legume flours are fractionated into separate protein fractions, LOX can act as undesirable "time-bombs" that can cause undesirable flavors on aging or storage. Compositions containing plant proteins (e.g., from ground plant seeds) can be subjected to purification to remove LOX using, for example, an affinity resin that binds to LOX and removes it from the protein sample. The affinity resin can be linoleic acid, linolenic acid, stearic acid, oleic acid, propyl gallate, or epigalloccatechin gallate attached to a solid support such as a bead or resin. See, e.g., WO2013138793. In addition, depending on the protein component of the food product, certain combinations of antioxidants and/or LOX inhibitors can be used as effective agents to minimize off-flavor or off-odor generation especially in the presence of fats and oils. Such compounds can include, for example, one or more of β-carotene, α-tocopherol, caffeic acid, propyl gallate, or epigallocatechin gallate.

In some embodiments, specific flavor compounds, such as those described in Tables 3, 8, 9, 12, 14, 16, or 17 can be isolated and purified from the flavor additive composition. These isolated and purified compounds can be used as an ingredient to create flavors useful to the food and fragrance industry.

A flavor additive composition can be in the form, of but not limited to, soup or stew bases, bouillon, e.g., powder or cubes, flavor packets, or seasoning packets or shakers. Such flavor additive compositions can be used to modulate the flavor and/or aroma profile for a variety of food products, and can be added to a consumable food product before, during, or after cooking of the food product.

Food Products

Food products containing one or more flavor precursors and one or more heme-containing proteins can be used as a base for formulating a variety of additional food products, including meat substitutes, soup bases, stew bases, snack foods, bouillon powders, bouillon cubes, flavor packets, or frozen food products. Meat substitutes can be formulated, for example, as hot dogs, burgers, ground meat, sausages, steaks, filets, roasts, breasts, thighs, wings, meatballs, meatloaf, bacon, strips, fingers, nuggets, cutlets, or cubes.

In addition, food products described herein can be used to modulate the taste and/or aroma profile of other food products (e.g., meat replicas, meat substitutes, tofu, mock duck or other gluten based vegetable product, textured vegetable protein such as textured soy protein, pork, fish, lamb, or poultry products such as chicken or turkey products) and can be applied to the other food product before or during cooking. Using the food products described herein can provide a particular meaty taste and smell, for example, the taste and smell of beef or bacon, to a non-meat product or to a poultry product.

Food products described herein can be packaged in various ways, including being sealed within individual packets or shakers, such that the composition can be sprinkled or spread on top of a food product before or during cooking.

Food products described herein can include additional ingredients including food-grade oils such as canola, corn, sunflower, soybean, olive or coconut oil, seasoning agents such as edible salts (e.g., sodium or potassium chloride) or herbs (e.g., rosemary, thyme, basil, sage, or mint), flavoring agents, proteins (e.g., soy protein isolate, wheat glutin, pea vicilin, and/or pea legumin), protein concentrates (e.g., soy protein concentrate), emulsifiers (e.g., lecithin), gelling agents (e.g., k-carrageenan or gelatin), fibers (e.g., bamboo filer or inulin), or minerals (e.g., iodine, zinc, and/or calcium).

Food products described herein also can include a natural coloring agent such as turmeric or beet juice, or an artificial coloring agent such as azo dyes, triphenylmethanes, xanthenes, quinines, indigoids, titanium dioxide, red #3, red #40, blue #1, or yellow #5.

Food products described herein also can include meat shelf life extenders such as carbon monoxide, nitrites, sodium metabisulfite, Bombal, vitamin E, rosemary extract, green tea extract, catechins and other anti-oxidants.

Food products described herein can be free of animal products (e.g., animal heme-containing proteins or other animal products).

In some embodiments, the food products can be soy-free, wheat-free, yeast-free, MSG-free, and/or free of protein hydrolysis products, and can taste meaty, highly savory, and without off odors or flavors.

Assessment of Food Products

Food products described herein can be assessed using trained human panelists. The evaluations can involve eyeing, feeling, chewing, and tasting of the product to judge product appearance, color, integrity, texture, flavor, and mouth feel, etc. Panelists can be served samples under red or under white light. Samples can be assigned random three-digit numbers and rotated in ballot position to prevent bias. Sensory judgments can be scaled for "acceptance" or "likeability" or use special terminology. For example, letter scales (A for excellent, B for good, C for poor) or number scales may be used (1 = dislike, 2 = fair, 3 = good; 4 = very good; 5=excellent). A scale can be used to rate the overall acceptability or quality of the food product or specific quality attributes such

beefiness, texture, and flavor. Panelists can be encouraged to rinse their mouths with water between samples, and given opportunity to comment on each sample.

In some embodiments, a food product described herein can be compared to another food product (e.g., meat or meat substitute) based upon olfactometer readings. In various embodiments, the olfactometer can be used to assess odor concentration and odor thresholds, odor suprathresholds with comparison to a reference gas, hedonic scale scores to determine the degree of appreciation, or relative intensity of odors.

In some embodiments, an olfactometer allows the training and automatic evaluation of expert panels. In some embodiments, a food product described herein causes similar or identical olfactometer readings. In some embodiments, the differences between flavors generated using the methods of the invention and meat are sufficiently small to be below the detection threshold of human perception.

In some embodiments, volatile chemicals identified using GCMS can be evaluated. For example, a human can rate the experience of smelling the chemical responsible for a certain peak. This information could be used to further refine the profile of flavor and aroma compounds produced using a heme-containing protein and one or more flavor precursors.

Characteristic flavor and fragrance components are mostly produced during the cooking process by chemical reactions molecules including amino acids, fats and sugars which are found in plants as well as meat. Therefore, in some embodiments, a food product is tested for similarity to meat during or after cooking. In some embodiments human ratings, human evaluation, olfactometer readings, or GCMS measurements, or combinations thereof, are used to create an olfactory map of the food product. Similarly, an olfactory map of the food product, for example, a meat replica, can be created. These maps can be compared to assess how similar the cooked food product is to meat.

In some embodiments, the olfactory map of the food product during or after cooking is similar to or indistinguishable from that of cooked or cooking meat. In some embodiments the similarity is sufficient to be beyond the detection threshold of human perception. The food product can be created so its characteristics are similar to a food

product after cooking, but the uncooked food product may have properties that are different from the predicate food product prior to cooking.

These results will demonstrate that the compositions of the invention are judged as acceptably equivalent to real meat products. Additionally, these results can demonstrate that compositions of the invention are preferred by panelist over other commercially available meat substitutes. So, in some embodiments the present invention provides for consumables that are significantly similar to traditional meats and are more meat like than previously known meat alternatives.

The invention will be further described in the following examples, which do not limit the scope of the invention described in the claims.

EXAMPLES

Example 1: Addition of heme-protein increases beefy qualities of replica burgers

Replica burgers containing the ingredients in Table 4 and the flavor precursors cysteine (10 mM), glutamic acid (10 mM), glucose (10 mM), and thiamine (1 mM) were prepared. Water was added to make up the balance. See, for example, U.S. Provisional Application No. 61/751,816, filed January 11, 2013. Control burgers were prepared as in Table 4 with precursors cysteine (10 mM), glutamic acid (10 mM), glucose (10 mM), and thiamine (1 mM) except LegH was omitted.

After cooking for 5 minutes at 150 C, the replica burgers were evaluated by a trained sensory panel. Panelists were served samples under red lights and each panelist individually evaluated the samples. Samples were assigned a random three-digit number and rotated in ballot position to prevent bias. Panelists were asked to evaluate cooked replica burger samples on multiple flavor, aroma, taste, texture and appearance attributes including but not limited to: beefiness, bloody quality, savory quality, and overall acceptability using a 7-point scale from 1=dislike extremely, to 7=like extremely. Panelists were encouraged to rinse their mouths with water between samples, and to fill out a survey to record their evaluation of each sample.

When replica burgers containing the LegH were compared to the control replica burgers without LegH, the samples containing LegH were rated significantly beefier, bloodier, more savory, and overall preferred compared to those that did not include LegH. See Table 5.

TABLE 4

Replica Burger Ingredients

Replica burger	**% precooked w/w**
Pea vicilin	3.86
Soy protein concentrate (SPC)	2.52
Bamboo fiber	0.34
NaCl	0.54
Pea legumin	2
Soy Protein Isolate (SPI) (Solae, St. Louis, MO)	4.68
Wheat gluten	4.68
Coconut oil	15
Soy lecithin	0.1
k-carrageenan	1
LegH	1

TABLE 5

Sensory evaluation of replica burger with Heme

Attribute	Beef 20/80	No Heme	1% Heme
Beefyness			
mean	5.33	1.30	3.20
STDEV	1.58	0.67	0.79
Bloody			
mean	4.00	1.10	2.78
STDEV	1.32	0.32	1.64
Savory			

mean	4.67	3.00	5.10
STDEV	1.22	1.63	0.57

Example 2: Replica burgers with a flavor precursor mixture taste beefy and bloody

Replica burgers containing a flavor precursor mixture of glucose, cysteine, thiamine, and glutamic acid and 1% LegH pre-cooked w/w (see Table 4) were prepared as described in Example 1, and evaluated by a trained sensory panel after the burgers were cooked for 5 minutes at 150 C. Control burgers included LegH and all other ingredients except for the flavor precursor mixture.

Panelists were asked to evaluate the samples overall improvement in taste and descriptively analyze each sample using a 5-point scale from 1=dislike extremely, to 5=like extremely. Panelists were encouraged to rinse their mouths with water between samples, and to fill out a survey to record their evaluation of each sample. The replicate burgers which included LegH and the flavor precursor mixture were described as having bouillon, gravy, meaty, bloody, savory, and beefy notes on taste, and were preferred to the same replica burger with LegH but no added flavor precursor mixture. See, Table 6

TABLE 6

Improvement of overall taste with precursors added to LegH burgers

	with precursors	without precursors
Average	3.5	1.8
STDV	0.6	0.5

Example 3: Replica burgers with flavor precursor mixture resulting in a bacon taste

Replica burgers (see Table 4) were cooked with different precursor mixes (see Table 7) and 1% LegH and evaluated by a trained sensory panel after the burgers were cooked for 5 minutes at 150 C. Control burgers contained LegH and all of the other ingredients except for the flavor precursors. Panelists were asked to evaluate each sample and descriptively analyze of each sample. 5-point scale from 1=dislike extremely, to 5=like extremely. Panelists were encouraged to rinse their mouths with water between

samples, and to fill out a survey to record their evaluation of each sample. A replica burger with a precursor mixture of 10 mM glucose, 10 mM ribose, 10 mM cysteine, 1 mM thiamine, 1 mM glutamic acid, 1 mM GMP, and LegH was described as having a bacon aroma and taste, and overall meatiness, savory quality, a very umami quality, a brothy quality, and slight beefy notes. See Table 7 for a summary of the flavor description for the various combinations of flavor precursors and heme-containing protein.

TABLE 7

Flavors generated by addition of precursors to LegH (1 %)

Precursor (concentration)						Flavor Description
	ribose (10 mM)	cysteine (10 mM)				some kind of cold-cut/sliced deli meat
	ribose (10 mM)	cysteine (10 mM)			IMP (2 mM)	bread crust with beef drippings, sweet, grassy, umami
	ribose (10 mM)	cysteine (10 mM)			lactic acid (1 mM)	bready, malty, browned, breadcrust
	ribose (10 mM)	cysteine (10 mM)			lysine (5 mM)	savory, beefy, little grassy, brothy, bread
	ribose (10 mM)	cysteine (10 mM)			alanine (5 mM)	savory, weak beefy, brothy, little metallic
	ribose (10 mM)	cysteine (10 mM)			I+G (2 mM)	savory, weak beefy, brothy, sweet
	ribose (10 mM)	cysteine (10 mM)			methionine	cooked potato
	ribose (10 mM)	cysteine (10 mM)		glutamic acid (5 mM)		little meaty, pretzel, brothy, savory, sweet, chocolate
glucose (10 mM)	ribose (10 mM)	cysteine (10 mM)	thiamine (2 mM)	glutamic acid (5 mM)		slight beefy, browned, grasssy,
glucose (10 mM)	ribose (10 mM)	cysteine (10 mM)	thiamine (2 mM)	glutamic acid (5 mM)	IMP (2 mM)	bacon, very umami, savory, brothy, slight beef
glucose (10 mM)		cysteine (10 mM)	thiamine (2 mM)	glutamic acid (5 mM)		beef jerky, bloody, meaty, brothy
glucose (10 mM)		cysteine (10 mM)	thiamine (2 mM)	glutamic acid (5 mM)	lactic acid (1 mM)	savory, beefy, bloody, meaty, savory, gravy

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glucose (10 mM)		cysteine (10 mM)	thiamine (2 mM)	glutamic acid (5 mM)	lysine (5 mM)	roast beef
glucose (10 mM)		cysteine (10 mM)	thiamine (2 mM)	glutamic acid (5 mM)	alanine (5 mM)	boiled beef, sweet
glucose (10 mM)		cysteine (10 mM)	thiamine (2 mM)	glutamic acid (5 mM)	I+G (2 mM)	beefy with a sulfury note
glucose (10 mM)		cysteine (10 mM)			I+G (2 mM)	sweet, malty, umami, meaty
glucose (10 mM)					I+G (2 mM)	savory, roast beef, grassy
glucose (10 mM)				glutamic acid (5 mM)		umami, savory, meaty, sweaty, fermented

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Example 4: Type of sugar modulates flavor compounds created in the presence of Hemeprotein

The addition of different sugars to flavor reaction mixtures containing a hemeprotein and one or more flavor precursor compounds resulted in distinct differences in the flavor compounds generated and the overall flavor profile. LegH heme protein at 1% pre-cooked w/w/ was mixed with cysteine (10 mM) and glucose (20 mM) at pH 6 in phosphate buffer to form a flavor reaction mixture and heated to 150 C for 3 minutes; this reaction created flavor compounds known to be present in meat; *see* Table 8. Similarly, a flavor reaction mixture made when LegH heme protein at 1% was mixed with cysteine (10 mM) and ribose (20 mM) at pH 6 and heated to 150 C for 3 minutes created flavor compounds known to be in meat; *see* Table 8.

The characteristic flavor and fragrance components were mostly produced during the cooking process when the flavor precursor molecules reacted with the heme-protein. Gas chromatography–mass spectrometry (GCMS) is a method that combines the features of gas-liquid chromatography and mass spectrometry to separate and identify different substances within a test sample. Samples were evaluated by GCMS to identify the flavor compounds generated after heating and also evaluated for their sensory profiles. Volatile chemicals were isolated from the head space around the flavor reactions. The profile of the volatile chemicals in the headspace around the flavor reaction mixtures is shown in Table 8. In particular, the use of ribose created some additional compounds as compared to glucose, as shown in Table 8.

Notably, the control mixtures of cysteine with ribose or glucose heated in the absence of the LegH heme-protein did not generate the same set of flavor compounds. The flavor reaction mixtures containing LegH also were evaluated by a blinded trained sensory panel, which described the samples with ribose as having beefy, savory, brothy, and gravy-like notes, and the samples with glucose as savory, bloody, metallic, raw meat, and bouillon-like.

TABLE 8

Flavor compounds generated with cysteine, LegH, and either glucose or ribose in the flavor reaction mixture.

LegH 1%		
Compounds created	cysteine (10 mM), glucose (20 mM)	cysteine (10 mM), ribose (20 mM)
benzaldehyde	X	X
2-butanone	X	X
dimethyl trisulfide	X	X
2-pentyl-furan	X	X
2-methyl-propanal	X	X
thiazole	X	X
butyrolactone	X	X
2-acetylthiazole	X	X
pentanal	X	X
3-methyl-butanal	X	X
methyl-thiirane	X	X
nonanal	X	X
heptanal	X	X
2,3-butanedione	X	X
1,3,5-cycloheptatriene	X	X
propyl-cyclopropane	X	X
2-hexyl-furan	X	X
butanal	X	X
2-methyl-butanal	X	X
2-ethyl-furan		X
2-octanone	X	X
propanal	X	X

trichloromethane	X	
2-methyl-furan	X	X
furan	X	X
pyrazine	X	X
thiophene	X	X
1,3-dimethyl-benzene	X	X
octane		X
octanal	X	X
thiazole	X	X
2-pentanone		X
furfural	X	X
2-nonanone	X	X
(Z)-2-heptenal	X	X
(E)-2-heptenal	X	X
1-octene	X	X
formic acid, heptyl ester	X	X
2-pentyl-thiophene		X
1-octen-3-one	X	X
3-pentyl-furan	X	X
2-propenal		X
(E)-2-tridecen-1-ol		X
benzene		X
(E)-4-octene		X
1-penten-3-one		X
4-penten-2-one	X	X
2-methyl-thiazole		X
methyl-pyrazine		X
trans-2-(2-pentenyl)furan		X
3-ethylcyclopentanone		X

pyrrole	X	X
2-thiophenecarboxaldehyde		X
3-thiophenecarboxaldehyde		X

Example 5: Heme-protein in the presence of thiamine affects the production of certain flavor compounds

The addition of thiamine in a flavor reaction mixtures with a heme protein and other flavor precursors affected the formation of 5-Thiazoleethanol, 4-methyl-furan, 3,3'-dithiobis[2-methyl-thiazole, and 4-methylthiazole. These compounds are known to be present in meat and have beefy, meaty taste notes.

Flavor reaction mixtures at pH 6 containing LegH (1%), cysteine (10 mM), thiamine (1 mM), either glucose or ribose (20 mM), and with or without glutamic acid (10 mM) were prepared and subsequently heated to 150 C for 3 minutes. These flavor reaction samples then were evaluated by GCMS for the flavor compounds generated and evaluated by a trained panel for their sensory profiles. Volatile chemicals were isolated from the head space around the flavor reactions. GCMS showed 4-methyl-5-thiazoleethanol, 3,3'-dithiobis[2-methyl]-furan, and 4-methylthiazole compounds were created by a mixture of LegH with thiamine, a sugar (either glucose or ribose), and cysteine. The same flavor reaction mixtures without thiamine did not generate these compounds; additionally these compounds were not generated when heme-proteins were not present in the flavor reaction mixtures.

The flavor reaction samples also were evaluated by a blinded trained sensory panel, which described the samples with the addition of thiamine as more complex in taste and more beefy, meaty, and savory.

Example 6: Heme-proteins with nucleotides controls particular flavor compound production.

The addition of inosine monophosphate and guanosine monophosphate in mixes with heme protein and other precursors controlled the formation of flavor compounds (E)-4-octene, 2-ethyl-furan, 2-pentanone, 2,3-butanedione, 2-methyl-thiazole, , methyl-

pyrazine, tridecane, (E)-2-octenal, 2-thiophenecarboxaldehyde, and 3-thiophenecarboxaldehyde. These compounds are known to be present in meat and have a beefy, meaty, buttery, and or savory flavor notes.

Reactions containing heme protein at 1% (LegH**)** with cysteine (10 mM), and glucose (20 mM), 1 mM IMP and 1 mM GMP, at pH 6.0 were prepared and heated to 150C for 3 minutes. Characteristic flavor and fragrance components were mostly produced during the cooking process where precursors reacting heme-protein. These samples were evaluated by GCMS for the flavor compounds generated and evaluated for the sensory experience. Volatile chemicals were isolated from the head space around the flavor reaction and identified using GCMS, creating a profile of the volatile chemicals in the headspace around the flavor reaction mixture. GCMS showed 4-octene, 2-ethyl furan, 2-pentanone, 2,3-butanedione, 2-methyl-thiazole, methyl-pyrazine, tridecane, 2-octenal, 2-thiophenecarboxaldehyde, 3-thiophenecarboxaldehyde compounds were created by a mixture of hemeprotein LegH with IMP, GMP, glucose, and cysteine. The same samples without IMP and GMP did not generate these compounds, additionally these compounds were also not created when heme-proteins were not present, just precursor molecules. Sensory evaluation by blinded trained panelist found the samples with the addition of inosine and guanosine as described as having more complexity in taste and more beefy, meaty, brothy and savory. Figure 2 shows the abundance of the novel flavor compounds created with heme protein at 1% was mixed in a reaction at pH 6, with cysteine (10 mM), and glucose (20 mM), IMP (1 mM) and GMP (1 mM), and detected by solid phase microextraction (SPME) and then detected by GCMS.

Example 7: Flavor generation with the addition of a particular organic acid

The addition of lactic acid in mixes with heme protein, ribose, and cysteine controlled the formation of the flavor compounds nonanal, thiazole, 2-acetylthiazole, and 8-methyl-1-undecene. These compounds are known to be present in meat.

Reactions containing heme protein at 1%, cysteine (10 mM), and ribose (20 mM), and lactic acid (1mM), pH 6.0, were prepared and heated to 150C for 3 minutes. Characteristic flavor and fragrance components were mostly produced during the cooking

process where precursors reacting heme-protein. These samples were evaluated by GCMS for the flavor compounds generated and evaluated for the sensory experience. Volatile chemicals were isolated from the head space around the flavor reaction and identified using GCMS, creating a profile of the generated compounds. Nonanal, thiazole, 2-acetylthiazole, and 8-methyl-1-undecene compounds were created by a mixture of LegH with lactic acid, ribose, and cysteine. The same samples without lactic acid did not generate these compounds, additionally these compounds were not created in the absence of heme-proteins.

Sensory evaluation by blinded trained panelist found the samples with the addition of lactic acid as described as beefy, savory, browned, bready, and having malty notes. The sample with everything but lactic acid rated lower in browned, bready and malty notes.

Example 8: Flavor generated with the addition of a particular amino acid.

The addition of lysine in mixes with heme protein ribose, and cysteine controlled the formation of flavor compounds dimethyl trisulfide, nonanal, 2-pentyl-thiophene, furfural, 2-nonenal, 1-octanol, 2-nonenal, thiazole, 2-acetylthiazole, phenylacetaldehyde, 2-acetylthiazole. These compounds are known to be present in meat and some have a beefy, meaty, and or savory flavor.

Reactions containing heme protein at 1%, cysteine (10 mM), and ribose (20 mM), and lysine (1mM), at pH 6.0, were prepared and heated to 150C for 3 minutes. These samples were evaluated by GCMS for the flavor compounds generated and evaluated for the sensory experience. Characteristic flavor and fragrance components were mostly produced during the cooking process where precursors could react with the heme-protein. These samples were evaluated by GCMS for the flavor compounds generated and evaluated for the sensory experience. Volatile chemicals were isolated from the head space around the flavor reaction. Dimethyl trisulfide, nonanal, 2-pentyl-thiophene, furfural, 2-nonenal, 1-octanol, 2-nonenal, thiazole, 2-acetylthiazole, phenylacetaldehyde, 2-acetylthiazole compounds were created by a mixture of LegH with lactic acid, ribose, and cysteine. The same samples without lactic acid did not generate these compounds,

additionally these compounds were not created when heme-proteins were not present, just precursor molecules. Sensory evaluation by blinded trained panelist found the samples with the addition of lysine as described as roast beefy, savory, and browned. The addition of lysine increased the roasted browned notes.

Example 9 - Flavor compound production by different Heme-proteins

The addition of different types of heme-proteins (LegH, Barley, *B. myoglobin*, or *A. aeolicus*) in flavor reaction mixtures containing one or more flavor precursor compounds results in many of the same key meat flavors, including but not limited to 2-pentyl-furan, 2,3-Butanedione, Thiophene, 2-methyl-thiazole, Pyrazine, Furan, Pyrrole, 2-methyl-furan and distinct differences in the flavor compounds, including but not limited to 2-pentyl-thiophene, Nonanal, 2-Nonanone, and 1-Octen-3-one. These differences in flavor compounds can change the overall taste profile. The different types of heme-protein were LegH, Barley, *B. myoglobin*, or *A. aeolicus* used at 1% w/w in a reaction mixed with cysteine (10 mM) and ribose (10 mM) at pH 6. The pre- reaction mixture was heated to 150 C for 3 minutes; this reaction created flavor compounds known to be present in meat; see Table 9. The characteristic flavor and fragrance components are mostly produced during the cooking process where the flavor precursor molecules react with the heme-protein. Samples were evaluated by GCMS to identify the flavor compounds generated after heating and also evaluated for their sensory profiles. Volatile chemicals were isolated from the head space around the flavor reactions. Table 9 shows the similarity and differences in volatile flavor compounds created by the different types of heme-proteins.

TABLE 9

Flavor compounds created by different heme-protein when heated with ribose and cysteine.

Name	LegH	Barley	*B. myoglobin*	*A. aeolicus*
Furan	x	x	x	x
Thiazole	x	x	x	x
benzaldehyde	x	x	x	x
2-acetylthiazole	x	x	x	x

2-methyl-propanal	x	x	x	x
furfural	x	x	x	x
2,3-butanedione	x	x	x	x
2-pentyl-furan	x	x	x	x
2-pentanone	x	x		
pyrazine	x	x	x	x
dimethyl trisulfide	x	x	x	x
3-methyl-butanal	x	x		x
2-methyl-thiazole	x	x	x	x
pentanal	x	x	x	x
1,3,5-cycloheptatriene	x	x	x	x
methacrolein	x	x	x	x
heptanal	x	x	x	x
2-methyl-butanal	x	x		x
isothiazole	x	x	x	x
thiophene	x	x	x	x
propanal	x	x	x	x
2-heptenal	x		x	x
methyl-pyrazine	x	x	x	x
1-octene	x		x	x
butanal	x	x	x	x
2-acetyl-propen-2-ol	x	x	x	x
pyrrole	x	x	x	x
2-methyl-furan	x	x	x	x
nonanal		x	x	x
2-propenal		x	x	x
2-decenal		x	x	x
2-nonanone		x		x
2-octanone		x	x	x
2-tridecen-1-ol,			x	x
2-octanone			x	
2-octenal			x	x
4-methyl-2-heptanone			x	x
octanal			x	x
2-undecenal				x
butyrolactone				x
1-octen-3-one				x
3-methylheptyl acetate				x

2-pentyl-thiophene				x

Example 10 - Generation of Meat flavors from different lipids

Several different samples including oils (canola oil or coconut oil), free fatty acids (FFA) (linoleic acid (C18:2), oleic acid (C18:1), stearic acid (C18:0), or myristic acid (C14:0)) and phospholipids (PL) (beef heart polar lipids extract, Biolipon95 (from Perimond), or NatCholinePC40 (from Perimond)) were tested for their ability to produce beefy flavor in the absence and in the presents of other precursors. Oils, FFAs, and PLs were added to 50 mM potassium phosphate buffer (PPB) pH 6.0 or a Maillard reaction mix (MRM) containing 50 mM potassium phosphate pH 6.0, 5 mM Cysteine, 10 mM Glucose, 0.1 mM Thiamine, and 0.1% (w/v) LegHemoglobin. Lipids in combination with MRM were designed to capture the cross reactions of lipid degradation and Maillard reaction productions while lipids in phosphate buffer functioned as a lipid control. The oils were added at 3% of the total 1 mL volume of solution while FFAs and PLs were added at 1% of the total 1 mL volumes. All samples were cooked at 150°C for 3 mins, cooled to 50°C and then analyzed using GCMS (SPME fiber sampling of headspace). After all samples were analyzed by GCMS the caps were removed and samples were smelled by a trained flavor scientist and aromas recorded.

Table 10

Legend showing components of each sample

Sample Name	Solution	Additives
MRM None	Maillard Reaction Mix	None
MRM_Linoelic Acid	Maillard Reaction Mix	1% linoleic acid
MRM_Oleic Acid	Maillard Reaction Mix	1% oleic acid
MRM_C14	Maillard Reaction Mix	1% C14:0 free fatty acid
MRM_C18	Maillard Reaction Mix	1% C18:0 free fatty acid
MRM_Canola	Maillard Reaction Mix	3% Canola Oil
MRM_Coconut	Maillard Reaction Mix	3% Coconut Oil
MRM_BeefHeart	Maillard Reaction Mix	1% Beef Heart Polar Lipids Extract
MRM_Biolipon95	Maillard Reaction Mix	1% Biolipon95 (emulsifier)

MRM_NatCholinePC40	Maillard Reaction Mix	1% NatCholinePC40 (emulsifier)
KPhos6_Linoelic Acid	PPB, pH 6	1% linoelic acid
KPhos6_Oleic Acid	PPB, pH 6	1% oleic acid
KPhos6_C14	PPB, pH 6	1% C14:0 free fatty acid
KPhos6_C18	PPB, pH 6	1% C18:0 free fatty acid
KPhos6_Canola	PPB pH 6	3% Canola Oil
KPhos6_Coconut	PPB, pH 6	3% Coconut Oil
KPhos6_BeefHeart	PPB, pH 6	1% Beef Heart Polar Lipids Extract
KPhos6_Biolipon95	PPB, pH 6	1% Biolipon95 (emulsifier)
KPhos6_NatCholinePC40	PPB, pH 6	1% NatCholinePC40 (emulsifier)

Table 11 contains the aroma descriptions and Table 12 contains the GCMS data from the most interesting samples analyzed. Many of the lipids introduced a "fatty" aroma to MRM that was otherwise absent. The combinations of Linoleic Acid or NatCholinePC40 in MRM produced the greatest abundance of fatty compounds suggesting that these lipids may improve the flavor perception of beef tallow. Linoleic Acid and NatCholinePC40 also showed high abundance of earthy-mushroom aromas. The addition of lipids to MRM significantly increased the abundance of "nutty & roasted" aromas. Less desirable "green" aroma compounds were most prominent in samples with unsaturated free fatty acids (linoleic acid or oleic acid) or phospholipids. In general, the addition of lipids significantly increased the number of target beef compounds made.

TABLE 11

Aroma descriptions of each sample after it was cooked.

Sample Names	Aroma Descriptions
MRM_Only	brothy, malty, beef stew
KPhos6_BeefHeart	**fatty, creamy, beef tallow, slight sweet, slight roasted nutty**
MRM_BeefHeart	**fatty, beef tallow, old meat, mushroom**
KPhos6_Biolipon95	fatty, fresh

MRM_Biolipon95	fatty, brothy, hay, malty green
KPhos6_NatCholinePC 40	**light fatty, fresh**
MRM_NatCholinePC40	**fatty, beef tallow, brothy**
K-Phos6_C14	light/faint plastic/waxy
MRM_C14	brothy, beefy, minty, fresh
K-Phos6_C18	**light/faint plastic/waxy**
MRM_C18	**beefy with cucumber &/or pepper aroma**
K-Phos6_Canola	fresh, cucumber
MRM_Canola	fatty, brothy, oil, roasted nuts
K-Phos6_Coconut	**nothing**
MRM_Coconut	**brothy, beefy, slight fatty, crackers**
K-Phos6_Oleic Acid	fresh, cucumber, camphorous/minty-like
MRM_Oleic Acid	herbal, plastic, slight cheesy, brothy
K-Phos6_Linoelic Acid	**light plastic**
MRM_Linoelic Acid	**fatty, light waxy, brothy, herbal**

Table 12

List of aromatic compounds found in Beef by GCMS and a chart showing which were detected in each lipid plus MRM sample.

Compounds in Beef	MRM only	MRM_BeefHeart	MRM_NatCholinePC40	MRM_Linoleic acid
(s)-isopropyl lactate	N	N	N	N
1-ethyl-5-methylcyclopentene	**Y**	**Y**	**Y**	**Y**
1-heptanol	N	**Y**	N	N
1-hepten-3-ol	N	**Y**	**Y**	**Y**
1-heptene	N	**Y**	**Y**	**Y**
2-methyl-1-heptene	N	N	N	N
1-hexanol	N	**Y**	**Y**	**Y**
2-ethyl-1-hexanol	N	N	N	N
1-nonanol	N	N	**Y**	N
1-nonene	N	**Y**	**Y**	N
1-octanol	N	**Y**	**Y**	N

1-octen-3-ol	N	**Y**	**Y**	**Y**
1-octen-3-one	**Y**	**Y**	**Y**	**Y**
1-octene	N	N	N	N
1-pentanol	N	**Y**	**Y**	**Y**
1-penten-3-ol	N	**Y**	**Y**	N
1-propanol	N	N	N	N
8-methyl-1-undecene	N	**Y**	**Y**	**Y**
1,3-hexadiene	N	N	N	**Y**
3-ethyl-2-methyl-1,3-hexadiene	N	**Y**	**Y**	**Y**
1,3-octadiene	**Y**	N	N	**Y**
1,3,5-cycloheptatriene	N	N	N	N
2,3-dihydro-5,6-dimethyl-1,4-dioxin	N	N	N	N
1,7-octadien-3-ol	N	**Y**	N	N
1h-pyrrole-2-carboxaldehyde	N	N	N	N
2-methyl-1H-pyrrole	N	N	N	N
2-acetyl-2-thiazoline	**Y**	N	N	N
2-acetylthiazole	**Y**	**Y**	**Y**	**Y**
2-butanone	N	**Y**	**Y**	**Y**
2-butenal	N	**Y**	**Y**	**Y**
2-ethyl-2-butenal	N	N	N	**Y**
3-methyl-2-butenal	N	N	**Y**	**Y**
3-methyl-2-cyclohexen-1-one	N	N	N	N
2-decanone	**Y**	**Y**	**Y**	N
(E)-2-decenal	N	N	N	N
(Z)-2-decenal	**Y**	**Y**	**Y**	**Y**
2-furanmethanol	N	N	N	N
2-heptanone	**Y**	**Y**	**Y**	**Y**
6-methyl-2-heptanone	N	N	**Y**	N
(E)-2-heptenal	N	**Y**	**Y**	**Y**
(Z)-2-heptenal	N	N	N	**Y**
(E)-2-hexenal	N	**Y**	**Y**	**Y**
2-ethyl-2-hexenal	N	N	N	N
2-methyl-2-heptene	**Y**	N	N	N
2-n-heptylfuran	**Y**	N	N	N

2-n-octylfuran	**Y**	**Y**	**Y**	N
2-nonanone	N	**Y**	**Y**	N
(E)-2-nonenal	**Y**	**Y**	**Y**	**Y**
(Z)-2-nonenal	N	N	N	**Y**
2-octanone	**Y**	**Y**	**Y**	**Y**
(Z)-2-octen-1-ol	**Y**	**Y**	**Y**	**Y**
(E)-2-octenal	N	**Y**	**Y**	**Y**
2-pentanone	N	**Y**	**Y**	N
1-propoxy-2-propanol	N	N	N	N
1-(acetyloxy)-2-propanone	**Y**	N	N	N
1-hydroxy-2-propanone	**Y**	N	N	N
2-propenal	N	N	N	**Y**
2-thiophenecarboxaldehyde	**Y**	**Y**	**Y**	**Y**
2-undecenal	N	**Y**	**Y**	**Y**
2,3-butanedione	N	N	N	**Y**
2,3-pentanedione	N	N	N	N
(E,E)-2,4-decadienal	N	**Y**	**Y**	**Y**
2,4-decadienal	N	N	N	**Y**
(E,E)-2,4-heptadienal	N	**Y**	**Y**	**Y**
(E,E)-2,4-nonadienal	N	**Y**	**Y**	**Y**
2,6-dimethylpyrazine	N	N	N	N
(E,Z)-2,6-nonadienal	N	N	**Y**	N
5-ethyldihydro-2(3H)-furanone	N	**Y**	**Y**	**Y**
5-methyl-2(3H)-furanone	N	N	N	N
dihydro-5-pentyl-2(3H)-furanone	N	N	**Y**	**Y**
dihydro-5-propyl-2(3H)-furanone	N	N	N	N
2(5H)-furanone	N	N	N	N
tetrahydro-6-methyl-2H-pyran-2-one	N	N	N	N
3-ethylcyclopentanone	N	**Y**	**Y**	**Y**
3-hexanone	N	N	N	N
3-methyl-2-thiophenecarboxaldehyde	N	N	N	N
3-octanone	**Y**	**Y**	N	**Y**
3-octen-2-one	N	**Y**	**Y**	**Y**

3-thiophenecarboxaldehyde	N	**Y**	**Y**	**Y**
(E,E)-3,5-octadien-2-one	N	N	**Y**	**Y**
dihydro-2-methyl-3(2H)-furanone	N	N	N	N
4-cyanocyclohexene	N	N	N	N
4-cyclopentene-1,3-dione	N	N	**Y**	N
4-decyne	N	Y	N	N
(Z)-4-heptenal	N	Y	**Y**	**Y**
4-methyloctanoic acid	N	N	N	N
(E)-4-octene	N	N	N	N
2,3-dihydro-3,5-dihydroxy-6-methyl-4(H)-pyran-4-one	**Y**	N	N	N
6-methyl-5-hepten-2-one	**Y**	N	N	N
acetaldehyde	N	N	N	**Y**
acetic acid	N	N	N	N
acetic acid ethenyl ester	**Y**	N	N	N
acetoin	**Y**	N	N	N
acetone	**Y**	N	N	**Y**
acetonitrile	N	N	N	**Y**
benzaldehyde	**Y**	**Y**	**Y**	**Y**
4-ethyl-benzaldehyde	N	**Y**	**Y**	N
benzene	**Y**	N	N	N
benzoic acid, hydrazide	**Y**	N	N	N
butanal	**Y**	N	N	**Y**
2-methyl-butanal	N	N	N	N
3-methyl-butanal	**Y**	N	N	N
butanoic acid	N	N	N	N
butyrolactone	**Y**	**Y**	N	**Y**
caprolactam	N	N	N	N
carbon disulfide	N	N	N	**Y**
1-ethyl-1-methyl-cyclopentane	**Y**	**Y**	**Y**	**Y**
propyl-cyclopropane	N	N	**Y**	**Y**
decanal	N	**Y**	**Y**	N
dihydro-3-(2H)-thiophenone	N	N	N	N
Dimethyl sulfide	**Y**	N	N	N
dimethyl sulfone	N	N	N	N
dimethyl trisulfide	**Y**	**Y**	N	N

ethanethiol	N	N	N	N
ethanol	N	N	N	**Y**
1-(1(H)-pyrrol-2-yl)-ethanone	N	N	N	N
1-(2-furanyl)-ethanone	N	N	N	N
ethosuximide	**Y**	N	N	N
formic acid, heptyl ester	**Y**	**Y**	N	N
furan	**Y**	N	N	**Y**
2-ethyl-furan	**Y**	N	N	N
2-hexyl-furan	**Y**	N	N	**Y**
2-methyl-furan	N	N	N	**Y**
2-pentyl-furan	N	**Y**	**Y**	**Y**
2-propyl-furan	N	N	**Y**	**Y**
3-methyl-furan	**Y**	N	N	N
3-pentyl-furan	**Y**	**Y**	**Y**	**Y**
furfural	N	**Y**	**Y**	**Y**
heptanal	N	**Y**	**Y**	**Y**
heptanoic acid	N	N	N	**Y**
2-methyl-hex-2-yn-4-one	N	N	N	N
hexanoic acid	N	N	N	**Y**
hydrogen sulfide	N	N	N	N
m-aminophenylacetylene	N	N	N	N
maleic anhydride	N	N	N	N
methacrolein	N	N	N	N
methanethiol	N	N	N	N
methyl ethanoate	N	N	N	N
methyl isobutyl ketone	**Y**	N	N	N
n-caproic acid vinyl ester	N	**Y**	**Y**	N
nonanal	N	**Y**	**Y**	**Y**
3-methyl-nonane	**Y**	N	N	N
nonanoic acid	**Y**	N	N	N
octanal	N	**Y**	**Y**	**Y**
octane	N	N	N	**Y**
octanoic acid	N	N	N	**Y**
oxalic acid, isobutyl pentyl ester	**Y**	N	N	N
p-cresol	N	N	N	N
pentanal	N	N	N	**Y**

pentanoic acid	**Y**	N	N	**Y**
4-ethyl-phenol	N	**Y**	**Y**	N
phenylacetaldehyde	**Y**	**Y**	**Y**	**Y**
(p-hydroxyphenyl)-phosphonic acid	**Y**	N	N	N
propanal	N	N	N	**Y**
2-methyl-propanal	N	N	N	N
propanoic acid	N	N	N	N
2-methyl-propanoic acid	**Y**	N	N	N
propanoic acid, ethenyl ester	N	N	N	N
pyrazine	N	**Y**	N	**Y**
2-ethyl-5-methyl-pyrazine	N	N	N	N
2-ethyl-6-methyl-pyrazine	N	N	N	N
2,3-dimethyl-pyrazine	N	N	N	N
2,5-dimethyl-pyrazine	N	N	N	N
3-ethyl-2,5-dimethyl-pyrazine	**Y**	N	N	N
ethyl-pyrazine	N	N	N	N
methyl-pyrazine	N	N	N	N
trimethyl-pyrazine	**Y**	N	N	N
pyridine	**Y**	N	**Y**	N
pyrrole	**Y**	**Y**	**Y**	**Y**
styrene	**Y**	N	**Y**	N
thiazole	**Y**	**Y**	**Y**	**Y**
methyl-thiirane	N	N	N	N
thiophene	N	N	N	**Y**
2-hexyl-thiophene	**Y**	N	**Y**	N
2-pentyl-thiophene	N	**Y**	N	N
trans-2-(2-pentenyl)furan	N	**Y**	**Y**	N
trans-3-nonen-2-one	N	**Y**	**Y**	**Y**
undecanoic acid	N	N	N	N
Total # of Compounds Detected:	54	63	66	76

In samples having fatty or creamy aromas, 2,4-decadienal, (E,E)-2,4-nonadienal, (E,E)-2,4-heptadienal, and/or (E,E)-2,4-decadienal were detected in the KPhos6_BeefHeart, MRM_BeefHeart, MRM_BioLipon95, MRM_NatCholinePC40,

Kphos6_Canola, MRM_Canola, KPhos6_Oleic Acid, KPhos6_Linoleic acid and MRM_Linoleic acid samples. For (E,E)-2,4-decadienal, the strongest signal intensity was in the MRM_NatCholinePC40 sample, followed by the MRM_Linoleic acid, KPhos6_Linoleic acid, MRM_BeefHeart, MRM_BioLipon95, KPhos6_BeefHeart, MRM_Oleic Acid, and KPhos6_Oleic Acid samples. For (E,E)-2,4-heptadienal, the strongest signal intensity was in the MRM_NatCholinePC40 sample followed by the MRM_Canola sample. (E,E)-2,4-heptadienal also was detected in the MRM_BioLipon95, MRM_BeefHeart, and MRM_Linoleic acid samples. For (E,E)-2,4-nonadienal, the strongest signal intensity was in the MRM_Canola and MRM_Linoleic acid samples. (E,E)-2,4-nonadienal also was detected in the Kphos6_Canola, MRM_NatCholinePC40, MRM_BioLipon95, MRM_BeefHeart, and KPhos6_Linoleic acid samples. For 2,4-decadienal, the strongest signal intensity was in the MRM_Linoleic acid sample. 2,4-decadienal also was detected in KPhos6_Linoleic acid, MRM_Canola, and KPhos6_Oleic Acid samples.

In samples having earthy or mushroom aromas, 3-octen-2-one, 1-octen-3-one, 3-octanone, and/or 1-octen-3-ol were detected in the KPhos6_BeefHeart, MRM_BeefHeart, Kphos_BioLipon95, MRM_BioLipon95, Kphos_NatCholinePC40, MRM_NatCholinePC40, MRM_Canola, KPhos6_Oleic Acid, MRM_Oleic Acid, KPhos6_Linoleic acid, and MRM_Linoleic acid samples. For 1-octen-3-ol, the strongest signal intensity was in the MRM_Linoleic acid sample, followed by MRM_NatCholinePC40, KPhos6_Linoleic acid, MRM_BeefHeart, KPhos6_BeefHeart, MRM_Canola, MRM_BioLipon95, KPhos6_Oleic Acid, and MRM_Oleic Acid samples. 3-octanone was detected in the MRM_Oleic Acid, KPhos6_Linoleic acid, and MRM_Linoleic acid samples. For 1-octen-3-one, the strongest signal intensity was in the MRM_Linoleic acid and MRM_BeefHeart samples, followed by KPhos6_Linoleic acid, MRM_NatCholinePC40, KPhos6_BeefHeart, MRM_BioLipon95, MRM_Oleic Acid, and KPhos6_Oleic Acid samples. For 3-octen-2-one, the strongest signal intensity was in the KPhos6_Linoleic acid sample, followed by MRM_Linoleic acid, MRM_NatCholinePC40, KPhos6_BeefHeart, KPhos6_Oleic Acid, MRM_Oleic Acid, MRM_BeefHeart, MRM_BioLipon95, MRM_Canola, Kphos_BioLipon95, and

Kphos_NatCholinePC40. Pyrazine was detected in the MRM_Coconut, MRM_C18, MRM_C14, and MRM_BioLipon95 samples.

In samples having a nutty and roasted aroma, thiazole and 2-acetylthiazole were the most abundant compounds detected, along with pyrazine, methyl pyrazine, trimethyl pyrazine, and 3-ethyl-2,5-dimethylpyrazine. 2-acetylthiazole was detected in all samples with MRM and most abundant in samples with MRM_Beefheat, MRM_biolipon95, MRM_Canola, and MRM_coconut. Thiazole was created in samples with MRM-Coconut, MRM_BeefHeat, MRM_Biolipon95, MRM_C14, MRM_C18, MRM_Canola, MRM_Oleic acid and MRM_Linoleic acid and MRM_NatCholinePC40. Pyrazine was present in the largest amount in samples with MRM-Coconut, followed by samples MRM_BeefHeat, MRM_Biolipon95, MRM_C14, MRM_C18, MRM_Canola having roughly equal amount, MRM_Oleic acid and MRM_Linoleic acid sample had even less. Methyl-pyrazine was present in MRM_Biolipon95 and MRM_Coconut. 3-ethyl-2,5-dimethyl- pyrazine and trimethyl- pyrazine, were present only without phospholipids in the MRM.

In samples having green, vegetable, or grass aromas, 1-heptanol, 1-hepten-3-ol, 1-hexanol, (E)-2-heptenal, (Z)-2-heptenal, (E)-2-hexenal, 2-pentyl-furan, and/or heptanal were detected in the KPhos6_BeefHeart, MRM_BeefHeart, Kphos_BioLipon95, MRM_BioLipon95, Kphos_NatCholinePC40, MRM_NatCholinePC40, Kphos_C14, MRM_C14, Kphos_C18, MRM_C18, MRM_Canola, MRM_Coconut, KPhos6_Oleic Acid, MRM_Oleic Acid, KPhos6_Linoleic acid, and MRM_Linoleic acid samples. For 2-pentyl-furan, the strongest signal intensity was in the KPhos6_BeefHeart sample, followed by the KPhos6_Linoleic acid, MRM_BioLipon95, MRM_Linoleic acid, MRM_BeefHeart, MRM_Oleic Acid, MRM_NatCholinePC40, MRM_Canola, KPhos6_Oleic Acid, and Kphos_NatCholinePC40 samples. For (E)-2-heptenal, the strongest signal intensity was in the MRM_BeefHeart, MRM_Canola, MRM_Oleic Acid, and KPhos6_Linoleic acid samples, followed by the KPhos6_Oleic Acid, MRM_BioLipon95, KPhos6_BeefHeart, MRM_Linoleic acid, MRM_NatCholinePC40, Kphos_BioLipon95, and Kphos_NatCholinePC40 samples. For (Z)-2-heptenal, the strongest signal intensity was in the MRM_Linoleic acid sample. MRM_Linoleic acid

also was detected in the KPhos6_Linoleic acid sample. For heptanal, the strongest signal intensity was in the MRM_Oleic Acid sample, followed by the KPhos6_Oleic Acid, MRM_C14, MRM_C18, MRM_Canola, MRM_BeefHeart, MRM_NatCholinePC40, MRM_Linoleic acid, and KPhos6_BeefHeart samples. For , (E)-2-hexenal, the strongest signal intensity was in the MRM_Linoleic acid sample, followed by the MRM_NatCholinePC40, KPhos6_Linoleic acid, and MRM_Oleic Acid samples.

Example 11 - Creation of beefy flavors using complex precursor mixtures

A formulation was prepared (the "magic mix," see Table 13 containing the estimated concentrations of amino acids, sugars, and other small molecules in beef based on their values reported in literature. The magic mix was tested for its ability to produce beefy flavors in the presence of LegHemoglobin (LegH). The magic mix and 1% w/v LegH were added to the meat replica, pH 6.0 (see Table 4) and baked in a convection oven for 7 minutes at 160°C. A control sample was prepared by adding 1% w/v LegH to the meat replica, pH 6.0 and baking in a convection oven for 7 minutes at 160°C.

The meat replica sample containing only LegH, was compared to the meat replica sample containing the magic mix and LegH by a sensory panel and GCMS analysis. Five tasters rated the flavored meat replicas for beefiness, bitterness, and levels of savory flavors, and off flavors. Each property was rated on a 7 point scale in which 7 was the highest amount of the specified property (e.g., a standard 80:20 ground beef would be rated 7 on the beefy scale). The Magic Mix flavor was rated one point higher in beefy character than the LegH only sample (FIG. 1).

To determine which chemical products were produced upon heating, a solution of Magic Mix was prepared with 1% w/v LegH at pH 6.0. The samples were cooked with shaking at 150°C for three minutes, then Solid Phase Micro Extraction (SPME) was performed for twelve minutes at 50°C to extract the volatile compounds above the headspace of the reaction. A search algorithm was used to analyze the retention time and mass fingerprint information of the volatile compounds and assign chemical names to peaks. Table 14 shows the compounds identified in both the Magic Mix + LegH (MM, average of two samples) and in the LegH alone in buffer (LegH, average of five samples)

samples. The compounds in Table 14 are listed in order of the retention time (R.T., in seconds), and are designated as having a zero peak area (0), or a small (S), medium (M), or large (L) average peak area. Hundreds of compounds were identified between the samples, many of which are characteristic of beefy aroma, including but not limited to 1,3-bis(1,1-dimethylethyl)-benzene, 2-methyl 3-furanthiol, and Bis(2-methyl-4,5-dihydro-3-furyl) disulfide, which increased in the samples containing the Magic Mix and LegH.

Table 13

Chemical entities added to the Magic Mix

Chemical entity	mM
Alanine	5.6
Arginine	0.6
Asparagine	0.8
Aspartate	0.8
Cysteine	0.8
Glutamic acid	3.4
Glutamine	0.7
Glycine	1.3
Histidine	0.6
Isoleucine	0.8
Leucine	0.8
Lysine	0.7
Methionine	0.7
Phenylalanine	0.6
Proline	0.9
Threonine	0.8
Tryptophan	0.5
Tyrosine	0.6

Valine	0.9
glucose	5.6
Ribose	6.7
Maltodextrin	5.0
Thiamine	0.5
GMP	0.24
IMP	0.6
Lactic acid	1.0
creatine	1.0
NaCl	10
KCl	10
Kphos pH 6.0	10

TABLE 14

Compounds identified with GC-MS analysis in samples with MM and LegH, or LegH alone (average of five samples)

R.T.(s)	Name	MM with LegH	LegH alone
248	acetaldehyde	L	S
256.3	carbon disulfide	L	S
264.3	dimethyl sulfide	S	0
265	oxalic acid, isobutyl pentyl ester	M	0
268.1	2,3,4-trimethyl-pentane	M	0
269.2	methanethiol	S	0
283.4	propanal	M	0
285.4	octane	M	0
287.1	furan	M	0
295.3	2-methyl-propanal	L	S
297.6	acetone	L	S
319.3	2-propenal	M	S
338.1	2-methyl-furan	M	S
342.1	butanal	L	S

344.2	2,4-dimethyl-1-heptene	M	0
346.3	methacrolein	M	0
357.4	methyl-thiirane	L	0
360.2	3-methyl-furan	S	0
363.7	butanone	L	S
368.9	2,3-dihydro-5-methyl-furan	M	S
376.4	2-methyl-butanal	L	M
381.1	3-methyl-butanal	L	M
390.6	isopropyl alcohol	0	S
399.6	ethanol	L	M
406.2	2-propenoic acid, methyl ester	M	0
408.2	benzene	S	0
414.4	methyl vinyl ketone	M	0
416.4	2,2,4,6,6-pentamethyl-heptane	M	0
422.6	2-ethyl-furan	S	0
438.4	2-ethylacrolein	M	0
449.9	2-pentanone	S	0
453.2	pentanal/2,3-butanedione	L	0
453.8	2,3-butanedione	L	M
472.8	4,7-dimethyl-undecane	M	S
485.9	2-methyl-pentanal	M	0
492.6	2-methyl-1-penten-1-one	S	0
496.6	(E)-3-penten-2-one	M	0
508.6	1-penten-3-one	M	0
510.6	trichloromethane	M	M
520.4	p-dithiane-2,5-diol	M	0
525.5	3-methyl-pentanal	M	0
535.1	(E)-5-decene	M	0
536.5	toluene	M	S
537.9	2-butenal	M	S
543.8	4-penten-2-one	M	0
550.8	methyl thiolacetate	M	0
683.7	p-xylene	S	0
727.4	dimethyl selenone	M	0
738.3	methyl isopropyl disulphide	M	0
755	2-heptanone	M	0
758.7	heptanal	L	0
781.9	1,3-diisopropoxy-1,3-dimethyl-1,3-	S	M

	disilacyclobutane		
789.4	3-methyl-2-butenal	M	0
793.4	4-methyl-2-heptanone	M	0
810.4	pyrazine	M	0
818.8	isothiazole	S	0
827.1	acetyl valeryl	M	0
831.8	2-pentyl-furan	L	0
851	2-methyl-thiazole	S	0
853.3	isothiocyanato-methane	S	0
870.9	thiazole	L	0
879.2	styrene	M	0
890.7	1-(methylthio)-propane	M	0
895.6	methyl-pyrazine	M	0
910.5	thiocyanic acid, methyl ester	S	0
918.6	4-methylthiazole	M	0
921.4	2-octanone	M	0
923.9	2-methyl-cyclopentanone	M	0
927.9	octanal	L	S
934.3	tridecane	M	0
948.8	trans-2-(2-pentenyl)furan	S	0
961.9	1-hydroxy-2-propanone	M	0
974.5	(E)-2-heptenal	M	0
987.4	5-methyl-1-undecene	M	0
993.8	2-hexyl-furan	M	0
1007.8	7-methyl-(E)-5-undecene	M	0
1024.1	2-methyl-5-(methylthio)-furan,	S	0
1058.6	2-butyl-1-decene	M	0
1079.3	dimethyl trisulfide	L	S
1085.3	2-nonanone	M	0
1093.2	nonanal	L	M
1142.3	1,3-bis(1,1-dimethylethyl)-benzene	M	0
1149.6	(E)-2-octenal	M	0
1164.5	1-heptanol	M	0
1193.5	methional	L	0
1198.8	acetic acid	M	S
1207.2	furfural	M	0
1242.1	2-decanone	M	0
1250.8	decanal	M	0

1265.2	1-decen-3-one	M	0
1283.3	pyrrole	M	0
1292.6	5-ethenyl-4-methyl-thiazole	M	0
1294.3	benzaldehyde	L	M
1303.7	2-n-octylfuran	M	0
1305.6	(E)-2-nonenal	M	0
1341.4	1-octanol	M	0
1361.1	2-methyl-1(H)-pyrrole	S	0
1391.7	2-undecanone	M	0
1401.2	(E)-2-octen-1-ol	M	0
1448	butyrolactone	S	S
1456.3	(E)-2-decenal	M	0
1462.4	phenylacetaldehyde	L	S
1466.3	2-acetylthiazole	L	0
1471.3	acetophenone	M	S
1475.4	1-nonanol	M	0
1487	methyl (methylthio)methyl disulfide	M	0
1497.1	5-(2-chloroethyl)-4-methylthiazole	L	0
1497.5	1-(ethylthio)-2-(methylthio)-buta-1,3-diene	L	S
1512	3-thiophenecarboxaldehyde	M	0
1518.8	2-nonen-4-one	M	0
1531.7	2-thiophenecarboxaldehyde	S	0
1543.9	dodecanal	M	0
1551.6	4-ethyl-2-methyl-pyrrole	S	0
1558.2	3-(methylthio)-propanenitrile	S	0
1561.2	3-decen-2-one	M	0
1613.1	bis(2-methyl-4,5-dihydro-3-furyl) disulfide	M	0
1615.6	1,10-undecadiene	M	0
1619.5	2-undecenal	S	0
1668.9	2-phenylpropenal	M	0
1692.3	(Z)-3-decen-1-ol, acetate	M	0
1733.1	3-phenyl-furan	S	0
1739.7	4-nitrophenyl 2-thiophenecarboxylic acid ester	S	0
1741.2	5-formyl-4-methylthiazole	M	0
1749.7	pentanoic acid, 2,2,4-trimethyl-3-hydroxy-, isobutyl ester	M	0
1765.5	benzyl alcohol	S	0

1774.2	pentanoic acid, 2,2,4-trimethyl-3-hydroxy-, isobutyl ester	S	0
1796.9	dodecanal	M	0
1806.1	(1-ethyl-1-propenyl)-benzene	S	0
1825.6	1-undecanol	M	S
1827.9	2-methyl-3-furanthiol	M	0
1828.3	2-methyl-3-(methylthio) furan	M	0
1836.1	4-chloro-2,6-bis(1,1-dimethylethyl)-phenol	S	0
1844.1	phenol	S	S
1845.3	[(methylsulfonyl)methyl]-benzene	S	0
1850.3	(e)-2-tridecen-1-ol	M	0
1859.9	1-heptyl-1,2,3,4-tetrahydro-4-methyl-naphthalene	S	0
1863.2	2,4-decadienal	S	0
1905.1	3,3'-dithiobis[2-methyl]-furan	M	0
1906.9	3,5-di-tert-butylbenzoic acid	S	0
1909.6	4-ethoxy-benzoic acid, ethyl ester	S	0
1921.5	3-(phenylmethyl)-2,5-piperazinedione	S	0
1944.5	9-octadecenal	M	0
1959.7	3,5-bis(1,1-dimethylethyl)-phenol	M	S
1968.4	4-methyl-5-thiazoleethanol	M	S
2007.8	1,1'-(1,2-cyclobutanediyl)bis-cis-benzene	S	0
2019.8	benzoic acid	S	S
2026.4	4-quinolinecarboxaldehyde	S	0
2027.8	m-aminophenylacetylene	M	0

Example 12 – Ferrous Chlorin Catalyzes Production of Meat-like flavor compounds

Fresh green spinach (10 lb) was added to 500 mL water and finely ground in a Vitamix blender to yield 2 L of green suspension. Acetone (8L) was added with mixing and the material was allowed to extract for 1 hour. The material was filtered through Whatman filter paper and the acetone was removed on a rotary evaporator (Buchi). To the residual green suspension (500 mL) was added 2 mL of 10 M HCl, causing the suspension to turn brown. To this was added 1g of $FeCl_2.4H_2O$ in 10 mL H_20. The

solution was shaken then left at 4°C for 16 hours. This suspension was extracted with diethyl ether (3x50mL) to give a bright green organic phase, the combined organics were washed with saturated sodium chloride solution, dried over sodium sulfate, filtered and evaporated to leave a black paste (1.1g). The pellet was dissolved in chloroform for fractionation.

Chlorophyll and Ferrous chlorin crude fractions were stored at -20°C. Crude extracts were fractionated by reverse-phase high-pressure liquid chromatography (RP-HPLC). HPLC conditions are outlined in Table 15. Both chlorophyll and ferrous chlorophyll were eluted from the column with a peak retention time of 7.6 minutes. Eluted material was collected from 7.3 – 8.0 minutes. Collected fractions were pooled and stored on ice. Collected fractions were re-chromatographed and showed a single peak with retention time 7.6 minutes. The desired fractions were pooled, then 10% sunflower oil was added, methanol was removed on a rotary evaporator (Buchi).

TABLE 15

HPLC conditions for purification of chlorophyll and ferrous chlorin from crude extract.

Sample:	Chlorophyll or Fe-chlorin (~2mg/mL in $CHCl_3$)
System:	Agilent 1100 with Chemstation
Column:	Zorbax Bonus-RP (4.6 x 250 mm, 5uM)
Mobile phase:	acetonitrile, methanol, ethyl acetate (60:20:20) isocratic flow
Temperature:	30°C
Flow Rate:	1.0 mL per minute
Injection volume:	0.05 mL

Preparation of Flavor Reaction Containing Ferrous Chlorin or Leghemoglobin

A solution of ferrous chlorophyll was mixed with the Magic Mix (Table 13) to a final concentration of 0.35% ferrous chlorin, 1% glycerol, 0.005% tween-20, 5% sunflower oil, 100 mM NaCl, 20 mM phosphate at pH 6. Leghemoglobin (0.35%) at pH 6 in phosphate buffer (20 mM), 100 mM NaCl, was mixed with the Magic Mix (Table

13), 1% glycerol, and 0.005% tween-20. The flavor reaction mixtures were heated to 150°C for 3 minutes; this reaction created flavor compounds known to be present in meat, created by hemoglobin and also created by ferrous chlorin; *see* Table 16.

The characteristic flavor and fragrance components were mostly produced during the cooking process when the flavor precursor molecules reacted with the heme-protein or the ferrous chlorophyll. Samples were evaluated by GCMS to identify the flavor compounds generated after heating. Volatile chemicals were isolated from the headspace around the flavor reactions. The profile of the volatile chemicals in the headspace around the flavor reaction mixtures that were similar between heme-protein and ferrous chlorin are shown in Table 16. Notabily, many of the compounds created by the ferrous chlorin are important in the flavor of meat.

TABLE 16

Flavor Compounds created by both Ferrous Chlorin and LegH with Magic Mix.

1-heptanol	acetone
1-hexanol	acetonitrile
1-octanol	benzaldehyde
1-octen-3-ol	butanal
1-octen-3-one	2-methyl-butanal
1-pentanol	dimethyl trisulfide
2-acetylthiazole	ethyl acetate
2-butenal	furan
3-methyl-2-butenal,	2-ethyl-furan
(Z)-2-decenal	2-hexyl furan
6-methyl-2-heptanone	2-pentyl-furan
(E)-2-heptenal	furfural
(E)-2-hexenal	heptanal
2-methyl-3-furanthiol	aminophenylacetylene
(E)-2-nonenal	methacrolein
(E)-2-octenal	methional
2-pentanone	octanal
1-hydroxy-2-propanone	octane
2-thiophenecarboxaldehyde	oxalic acid, diallyl ester
2-undecenal	2,3-butanedione

3-methyl-3-buten-2-one	2-methyl-propanal
3-thiophenecarboxaldehyde	pyrazine
(E)-4-octene,	2,3-dimethyl-pyrazine
methyl-pyrazine	2,5-dimethyl-pyrazine
thiazole	

Example 13 - Flavor creation by immobilized Hemin

Preparation of hemin linked CM Sepharose.

200 mg of bovine hemin (Sigma Aldrich) was loaded into a scintillation vial. A small magnetic stir bar, 800 μL acetonitrile, 64 μL 4-methylmorpholine, and 71 mg of N-hydroxysuccinimide were added in that order. The vial was placed in an ice bath and chilled then 118 mg of N-(3-dimethylaminopropyl)-N'-ethyl-carbodiimide hydrochloride was added with stirring, followed by 845 μL of Jeffamine ED900. This was stirred while allowing the black mixture to warm to ambient temperature. Chloroform (10 mL) was added to the mixture followed by water (4 mL). A flashlight was used to distinguish between organic and aqueous layers since both were black and the organic layer was pipetted off and concentrated to a dark black oil. The oil was dissolved in a 4:1 mixture of acetonitrile and ethanol to make an approximately 10% strength solution that was inky black in color.

2 mL of water swelled and equilibrated CM Sepharose was equilibrated in a BioRad minicolumn with 3 volumes of acetonitrile. The resin was resuspended in 1 mL acetonitrile and pipetted into a scintillation vial. This was followed with 44 microliters 4-methylmorpholine, 23 mg N-hydroxysuccinimide, and 39 mg of solid N-(3-dimethylaminopropyl)-N'-ethyl-carbodiimide hydrochloride. The mixture was vortexed vigorously and then shaken for three hours. To this white solid was added 570 microliters of inky black 20% strength hemin coupled diamine. The black solid was vortexed and shaken for an hour. The slurry strongly resembled Turkish coffee. The mixture was poured into a BioRad minicolumn and filtered, washed with acetonitrile until what came out no longer resembled espresso, then switched to deionized water, and finally 20 mM pH 9 sodium carbonate buffer. The black solid was washed until the effluent ran clear and then resuspended in 2 mL of buffer for storage until use.

Flavor reaction

The flavor reaction was created with heme protein (equine myoglobin -Sigma) at 0.35% in a phosphate buffer (20 mM) at pH 6.0 with 100 mM NaCl , this was mixed with Magic Mix (Table 13). Another flavor reaction was created with Immobilized Hemin at 0.35% in a phosphate buffer (20 mM) at pH 6.0 with 100 mM NaCl, this was mixed with Magic Mix (Table 13). The flavor reaction mixtures were heated to 150°C for 3 minutes; this reaction created flavor compounds known to be present in meat.

The characteristic flavor and fragrance components were mostly produced during the cooking process when the flavor precursor molecules reacted with the Heme-protein or the immobilized Hemin. Samples were evaluated by GCMS to identify the flavor compounds generated after heating. Volatile chemicals were isolated from the headspace around the flavor reactions. As can be seen in Table 17, immobilized hemin catalyzed production of compounds similar to those whose production was catalyzed by myoglobin free in solution. Notably, the profiles of flavor compounds, measured by GCMS, produced by cooking mixtures containing the immobilized hemin and the heme-protein, respectively, were very similar.

TABLE 17

Flavor compounds produced by cooking mixtures containing either myoglobin free in solution or hemin coupled to a solid support

Flavor compound	myoglobin	hemin-linker-resin
2-methyl-5-(methylthio)-thiophene	Low	
dihydro-5-propyl-2(3H)-furanone	Low	
octane	Low	
pyrrole	Low	Low
methanethiol	Low	Low
2-thiophenecarboxaldehyde	Low	Low
methyl-pyrazine	Low	Low
1-hydroxy-2-propanone	Low	Low
propanal	Low	Low

thiophene	Low	medium
pyridine	Low	Low
2-methyl-furan	Low	medium
oxalic acid, butyl propyl ester	Low	Low
pyrazine	medium	Low
oxalic acid, diallyl ester	medium	medium
2-butenal	medium	large
furfural	medium	medium
nonanal	medium	medium
2-ethyl-furan	medium	Low
ethanol	medium	very large
tert-butanol	medium	
3,3'-dithiobis[2-methyl]-furan	medium	medium
m-aminophenylacetylene	medium	medium
2,5-dihydro-3,4-dimethyl-furan	medium	medium
2-acetylthiazole	medium	medium
cyclohexane	medium	
ethyl tert-butyl ether	medium	
carbon disulfide	medium	medium
thiazole	medium	medium
acetonitrile	medium	large
2-pentyl-furan	medium	Low
3-thiophenecarboxaldehyde	medium	medium
2-methyl-butanal	medium	medium
thiazole	medium	large
2-methyl-3-furanthiol	large	large
2-propenal	large	large
3-methyl-2-butenal	large	medium
2-methyl-3-(methylthio) furan	large	large
ethyl acetate	large	medium
methacrolein	large	medium
methyl-thiirane	large	large
methional	large	large
methyl alcohol	large	medium
2-butanone	large	Low
2,3-butanedione	large	medium
acetone	large	large
furan	large	medium

benzaldehyde	large	medium
methyl thiolacetate	large	medium
acetaldehyde	very large	very large
2-methyl-propanal	very large	very large
dimethyl trisulfide	very large	very large
3-methyl-butanal	very large	very large
propyl-cyclopropane		medium
(E)-2-octenal		medium
2-n-propylaziridine		medium
thiirane		medium
ethyl formate		medium
methyl vinyl ketone		medium
2-propenoic acid, ethyl ester		medium
1-nonanol		large
1-octene		large
1-heptanol		large
1-dodecene		large
phorone		very large

Example 14. The combination of precursors with Heme protein drives flavor reactions.

Three samples were compared: precursor mix alone, 1% heme protein alone, and precursor mix with 1% heme. The precursor mix was made of glucose (20 mM), ribose (20 mM), cysteine (10 mM), thiamine (1 mM), and glutamic acid (1 mM). Reactions were all at pH 6.0, prepared and heated to 150°C for 3 minutes. These three samples were run in duplicate. These samples were evaluated by GCMS for the flavor compounds generated. Characteristic flavor and fragrance components were mostly produced during the cooking process where precursors could react with the heme-protein. These samples were evaluated by GCMS for the flavor compounds generated and evaluated for the sensory experience. Volatile chemicals were isolated from the head space around the flavor reaction. The flavor compounds created in each sample is indicated in Table 18. As shown most of the flavor molecules were created on when the precursors are combined with the heme protein.

Table 18

Flavor molecules created by the combination of LegH and precursor mix.

Compound	Precursor mix	LegH	Precursor mix + Leg H
carbon disulfide	medium	medium	high
isopropyl alcohol	medium	medium	low
2-methyl-furan	low		low
butanal	low		medium
thiophene	low		low
2,3-butanedione	low	low	high
furan	low		medium
2,4-dimethyl-1-heptene		high	high
acetone		high	high
dimethyl trisulfide		medium	medium
2-methyl-heptane		medium	medium
2-pentanone		medium	
pentanal		medium	medium
2-pentyl-furan		medium	medium
2-methyl-propanal		low	high
2-acetatyl-1-propene		low	low
2-methyl-butanal		low	medium
1,3-dimethyl-benzene		low	low
octane		low	low
benzene		low	low
benzaldehyde			very high
2-butanone			very high
furfural			very high
thiazole			high
nonanal			high
thiazole			high
2-acetylthiazole			medium
3-methyl-butanal			medium
(Z)-2-heptenal			medium
heptanal			medium
methyl-thiirane			medium
3-ethyl-pentane			medium
phenylacetaldehyde			medium
2-hexyl-furan			medium

2-nonanone			medium
propanal			medium
pyrazine			medium
(Z)-2-heptenal			medium
2-methyl-1-heptene			medium
2-ethyl-furan			medium
octanal			medium
(E)-4-octene			low
(E)-2-octenal			low
2-methyl-thiazole			low
2-propenal			low
1-octen-3-one			low
1-octene			low
2-octanone			low
dimethyl sulfide			low
3-pentyl-furan			low
2-n-octylfuran			low
2-pentyl-thiophene			low

OTHER EMBODIMENTS

It is to be understood that while the invention has been described in conjunction with the detailed description thereof, the foregoing description is intended to illustrate and not limit the scope of the invention, which is defined by the scope of the appended claims. Other aspects, advantages, and modifications are within the scope of the following claims.

WHAT IS CLAIMED IS:

1. A food product comprising:

a) an isolated and purified heme-containing protein or a highly conjugated heterocyclic ring complexed to iron ion; and

b) one or more flavor precursor molecules selected from the group consisting of glucose, fructose, ribose, arabinose, glucose-6-phosphate, fructose 6-phosphate, fructose 1,6-diphosphate, inositol, maltose, sucrose, maltodextrin, glycogen, nucleotide-bound sugars, molasses, a phospholipid, a lecithin, inosine, inosine monophosphate (IMP), guanosine monophosphate (GMP), pyrazine, adenosine monophosphate (AMP), lactic acid, succinic acid, glycolic acid, thiamine, creatine, pyrophosphate, vegetable oil, algal oil, sunflower oil, corn oil, soybean oil, palm fruit oil, palm kernel oil, safflower oil, flaxseed oil, rice bran oil, cottonseed oil, olive oil, sunflower oil, canola oil, flaxseed oil, coconut oil, mango oil, linoleic acid, a free fatty acid, cysteine, methionine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine, proline, serine, tyrosine, glutathione, an amino acid derivative, a protein hydrolysate, a malt extract, a yeast extract, and a peptone.

2. The food product of claim 1, wherein the highly conjugated heterocyclic ring complexed to an iron ion is selected from the group consisting of porphyrin, porphyrinogen, corrin, corrinoid, chlorin, bacteriochorophyll, corphin, chlorophyllin, bacteriochlorin, and isobacteriochlorin moiety complexed to iron ion.

3. The food product of claim 2, wherein the highly conjugated heterocyclic ring complexed to iron is ferrous chlorin.

4. The food product of claim 1, wherein the heme-containing protein is a plant heme-containing protein, an animal heme-containing protein, a yeast or filamentous fungus heme-containing protein, or a bacterial or archeal heme-containing protein.

5. The food product of claim 1, wherein the heme-containing protein is a non-symbiotic hemoglobin.

6. The food product of claim 1, wherein the heme-containing protein is a leghemoglobin.

7. The food product of claim 6, wherein the leghemoglobin is from soybean, alfalfa, lupin, fava bean, pea, cow pea, or lupine.

8. The food product of claim 1, wherein the isolated and purified heme-containing protein is recombinantly produced, chemically synthesized, or translated *in vitro*.

9. The food product of claim 1, wherein the heme-containing protein comprises an amino acid sequence having at least 80% sequence identity to a polypeptide set forth in SEQ ID NO:1.

10. The food product of claim 1, wherein the heme-containing protein is selected from the group consisting of androglobin, cytoglobin, globin E, globin X, globin Y, hemoglobin, myoglobin, erythrocruorin, beta hemoglobin, alpha hemoglobin, protoglobin, cyanoglobin, cytoglobin, histoglobin, neuroglobins, chlorocruorin, a truncated hemoglobin, truncated 2/2 globin, hemoglobin 3, cytochrome, and peroxidase.

11. The food product of claim 1, wherein the food product comprises two to fifty flavor precursors.

12. The food product of claim 1, wherein the food product comprises two to thirty-five flavor precursors.

13. The food product of claim 1, wherein the food product comprises two to ten flavor precursors.

14. The food product of claim 1, wherein the food product comprises two to six flavor precursors.

15. The food product of claim 1, further comprising a food-grade oil, a seasoning agent, a flavoring agent, a protein, a protein concentrate, an emulsifier, a gelling agent, or a fiber.

16. The food product of claim 1, wherein the food product is a meat substitute.

17. The food product of claim 1, wherein the food product is a broth, soup base, stew base, snack food, bouillon powder, bouillon cube, or a flavor packet.

18. The food product of claim 1, wherein the food product is a frozen food product.

19. The food product of claim 1, wherein the food product is sealed within a packet or shaker.

20. The food product of claim 1, wherein the food product is free of animal products.

21. A method of making a food product, the method comprising combining an isolated and purified heme-containing protein or a highly conjugated heterocyclic ring complexed to iron ion with one or more flavor precursor molecules to form a mixture, the one or more flavor precursor molecules selected from the group consisting of glucose, fructose, ribose, arabinose, glucose-6-phosphate, fructose 6-phosphate, fructose 1,6-diphosphate, inositol, maltose, sucrose, maltodextrin, glycogen, nucleotide-bound sugars, molasses, a phospholipid, a lecithin, inosine, IMP, GMP, pyrazine, AMP, lactic acid, succinic acid, glycolic acid, thiamine, creatine, pyrophosphate, sunflower oil, canola oil, flaxseed oil, coconut oil, mango oil, a free fatty acid, cysteine, methionine, isoleucine, leucine, lysine, phenylalanine, threonine, tryptophan, valine, arginine, histidine, alanine, asparagine, aspartate, glutamate, glutamine, glycine, proline, serine, tyrosine, glutathione, an amino acid derivative, a hydrolyzed protein, a malt extract, a yeast extract, and a peptone; and heating the mixture.

22. The method of claim 21, wherein the highly conjugated heterocyclic ring complexed to an iron ion is selected from the group consisting of porphyrin, porphyrinogen, corrin, corrinoid, chlorin, bacteriochorophyll, corphin, chlorophyllin, bacteriochlorin, and isobacteriochlorin moiety complexed to iron ion.

23. The method of claim 21, wherein the highly conjugated heterocyclic ring complexed to iron is ferrous chlorin.

24. The method of claim 21, wherein the heme-containing protein is a plant heme-containing protein, an animal heme-containing protein, a yeast or filamentous fungus heme-containing protein, or a bacterial or archeal heme-containing protein.

25. The method of claim 21, wherein the heme-containing protein is a non-symbiotic hemoglobin.

26. The method of claim 21, wherein the heme-containing protein is a leghemoglobin.

27. The method of claim 26, wherein the leghemoglobin is from soybean, alfalfa, lupin, fava bean, pea, cow pea, or lupine.

28. The method of claim 21, wherein the heme-containing protein is recombinantly produced, chemically synthesized, or transcribed and translated *in vitro*.

29. The method of claim 21, wherein the heme-containing protein comprises an amino acid sequence having at least 80% sequence identity to a polypeptide set forth in SEQ ID NO:1.

30. The method of claim 21, wherein the heme-containing protein is selected from the group consisting of androglobin, cytoglobin, globin E, globin X, globin Y, hemoglobin, myoglobin, erythrocruorin, beta hemoglobin, alpha hemoglobin, protoglobin, cyanoglobin, cytoglobin, histoglobin, neuroglobins, chlorocruorin, a truncated hemoglobin, truncated 2/2 globin, hemoglobin 3, cytochrome, and peroxidase.

ABSTRACT

This document relates to food products containing highly conjugated heterocyclic rings complexed to an iron ion and one or more flavor precursors, and using such food products to modulate the flavor and/or aroma profile of other foods. The food products described herein can be prepared in various ways and can be formulated to be free of animal products.

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FIG. 1

SEQ ID NO:1 *Vigna radiata*
MTTTLERGFTEEQEALVVKSWNVMKKNSGELGLKFFLKIFEIAPSAQKLFSFLRDSTVPLEQNPK
LKPHAVSVFVMTCDSAVQLRKAGKVTVRESNLKKLGATHFRTGVANEHFEVTKFALLETIKEAVP
EMWSPAMKNAWGEAYDQLVDAIKYEMKPPSS

SEQ ID NO:2 *Methylacidiphilum infernorum*
MIDQKEKELIKESWKRIEPNKNEIGLLFYANLFKEEPTVSVLFQNPISSQSRKLMQVLGILVQGI
DNLEGLIPTLQDLGRRHKQYGVVDSHYPLVGDCLLKSIQEYLGQGFTEEAKAAWTKVYGIAAQVM
TAE

SEQ ID NO:3 *Aquifex aeolicus*
MLSEETIRVIKSTVPLLKEHGTEITARMYELLFSKYPKTKELFAGASEEQPKKLANAIIAYATYI
DRLEELDNAISTIARSHVRRNVKPEHYPLVKECLLQAIEEVLNPGEEVLKAWEEAYDFLAKTLIT
LEKKLYSQP

SEQ ID NO:4 *Glycine max*
MGAFTEKQEALVSSSFEAFKANIPQYSVVFYTSILEKAPAAKDLFSFLSNGVDPSNPKLTGHAEK
LFGLVRDSAGQLKANGTVVADAALGSIHAQKAITDPQFVVVKEALLKTIKEAVGDKWSDELSSAW
EVAYDELAAAIKKAF

SEQ ID NO:5 *Hordeum vulgare*
MSAAEGAVVFSEEKEALVLKSWAIMKKDSANLGLRFFLKIFEIAPSARQMFPFLRDSDVPLETNP
KLKTHAVSVFVMTCEAAAQLRKAGKITVRETTLKRLGGTHLKYGVADGHFEVTRFALLETIKEAL
PADMWGPEMRNAWGEAYDQLVAAIKQEMKPAE

SEQ ID NO:6 *Magnaporthe oryzae*
MDGAVRLDWTGLDLTGHEIHDGVPIASRVQVMVSFPLFKDQHIIMSSKESPSRKSSTIGQSTRNG
SCQADTQKGQLPPVGEKPKPVKENPMKKLKEMSQRPLPTQHGDGTYPTEKKLTGIGEDLKHIRGY
DVKTLLAMVKSKLKGEKLKDDKTMLMERVMQLVARLPTESKKRAELTDSLINELWESLDHPPLNY
LGPEHSYRTPDGSYNHPFNPQLGAAGSRYARSVIPTVTPPGALPDPGLIFDSIMGRTPNSYRKHP
NNVSSILWYWATIIIHDIFWTDPRDINTNKSSSYLDLAPLYGNSQEMQDSIRTFKDGRMKPDCYA
DKRLAGMPPGVSVLLIMFNRFHNHVAENLALINEGGRFNKPSDLLEGEAREAAWKKYDNDLFQVA
RLVTSGLYINITLVDYVRNIVNLNRVDTTWTLDPRQDAGAHVGTADGAERGTGNAVSAEFNLCYR
WHSCISEKDSKFVEAQFQNIFGKPASEVRPDEMWKGFAKMEQNTPADPGQRTFGGFKRGPDGKFD
DDDLVRCISEAVEDVAGAFGARNVPQAMKVVETMGIIQGRKWNVAGLNEFRKHFHLKPYSTFEDI
NSDPGVAEALRRLYDHPDNVELYPGLVAEEDKQPMVPGVGIAPTYTISRVVLSDAVCLVRGDRFY
TTDFTPRNLTNWGYKEVDYDLSVNHGCVFYKLFIRAFPNHFKQNSVYAHYPMVVPSENKRILEAL
GRADLFDFEAPKYIPPRVNITSYGGAEYILETQEKYKVTWHEGLGFLMGEGGLKFMLSGDDPLHA
QQRKCMAAQLYKDGWTEAVKAFYAGMMEELLVSKSYFLGNNKHRHVDIIRDVGNMVHVHFASQVF
GLPLKTAKNPTGVFTEQEMYGILAAIFTTIFFDLDPSKSFPLRTKTREVCQKLAKLVEANVKLIN
KIPWSRGMFVGKPAKDEPLSIYGKTMIKGLKAHGLSDYDIAWSHVVPTSGAMVPNQAQVFAQAVD
YYLSPAGMHYIPEIHMVALQPSTPETDALLLGYAMEGIRLAGTFGSYREAAVDDVVKEDNGRQVP
VKAGDRVFVSFVDAARDPKHFPDPEVVNPRRPAKKYIHYGVGPHACLGRDASQIAITEMFRCLFR
RRNVRRVPGPQGELKKVPRPGGFYVYMREDWGGLFPFPVTMRVMWDDE

SEQ ID NO:7 *Fusarium oxysporum*
MKGSATLAFALVQFSAASQLVWPSKWDEVEDLLYMQGGFNKRGFADALRTCEFGSNVPGTQNTAE
WLRTAFHDAITHDAKAGTGGLDASIYWESSRPENPGKAFNNTFGFFSGFHNPRATASDLTALGTV
LAVGACNGPRIPFRAGRIDAYKAGPAGVPEPSTNLKDTFAAFTKAGFTKEEMTAMVACGHAIGGV

FIG. 1-CONT.

HSVDFPEIVGIKADPNNDTNVPFQKDVSSFHNGIVTEYLAGTSKNPLVASKNATFHSDKRIFDND
KATMKKLSTKAGFNSMCADILTRMIDTVPKSVQLTPVLEAYDVRPYITELSLNNKNKIHFTGSVR
VRITNNIRDNNDLAINLIYVGRDGKKVTVPTQQVTFQGGTSFGAGEVFANFEFDTTMDAKNGITK
FFIQEVKPSTKATVTHDNQKTGGYKVDDTVLYQLQQSCAVLEKLPNAPLVVTAMVRDARAKDALT
LRVAHKKPVKGSIVPRFQTAITNFKATGKKSSGYTGFQAKTMFEEQSTYFDIVLGGSPASGVQFL
TSQAMPSQCS

SEQ ID NO:8 *Fusarium graminearum*
MASATRQFARAATRATRNGFAIAPRQVIRQQGRRYYSSEPAQKSSSAWIWLTGAAVAGGAGYYFY
GNSASSATAKVFNPSKEDYQKVYNEIAARLEEKDDYDDGSYGPVLVRLAWHASGTYDKETGTGGS
NGATMRFAPESDHGANAGLAAARDFLQPVKEKFPWITYSDLWILAGVCAIQEMLGPAIPYRPGRS
DRDVSGCTPDGRLPDASKRQDHLRGIFGRMGFNDQEIVALSGAHALGRCHTDRSGYSGPWTFSPT
VLTNDYFRLLVEEKWQWKKWNGPAQYEDKSTKSLMMLPSDIALIEDKKFKPWVEKYAKDNDAFFK
DFSNVVLRLFELGVPFAQGTENQRWTFKPTHQE

SEQ ID NO: 9 *Chlamydomonas eugametos*
MSLFAKLGGREAVEAAVDKFYNKIVADPTVSTYFSNTDMKVQRSKQFAFLAYALGGASEWKGKDM
RTAHKDLVPHLSDVHFQAVARHLSDTLTELGVPPEDITDAMAVVASTRTEVLNMPQQ

SEQ ID NO:10 *Tetrahymena pyriformis*
MNKPQTIYEKLGGENAMKAAVPLFYKKVLADERVKHFFKNTDMDHQTKQQTDFLTMLLGGPNHYK
GKNMTEAHKGMNLQNLHFDAIIENLAATLKELGVTDAVINEAAKVIEHTRKDMLGK

SEQ ID NO:11 *Paramecium caudatum*
MSLFEQLGGQAAVQAVTAQFYANIQADATVATFFNGIDMPNQTNKTAAFLCAALGGPNAWTGRNL
KEVHANMGVSNAQFTTVIGHLRSALTGAGVAAALVEQTVAVAETVRGDVVTV

SEQ ID NO:12 *Aspergillus niger*
MPLTPEQIKIIKATVPVLQEYGTKITTAFYMNMSTVHPELNAVFNTANQVKGHQARALAGALFAY
ASHIDDLGALGPAVELICNKHASLYIQADEYKIVGKYLLEAMKEVLGDACTDDILDAWGAAYWAL
ADIMINREAALYKQSQG

SEQ ID NO:13 *Zea mays*
MALAEADDGAVVFGEEQEALVLKSWAVMKKDAANLGLRFFLKVFEIAPSAEQMFSFLRDSDVPLE
KNPKLKTHAMSVFVMTCEAAAQLRKAGKVTVRETTLKRLGATHLRYGVADGHFEVTGFALLETIK
EALPADMWSLEMKKAWAEAYSQLVAAIKREMKPDA

SEQ ID NO:14 *Oryza sativa subsp. japonica*
MALVEGNNGVSGGAVSFSEEQEALVLKSWAIMKKDSANIGLRFFLKIFEVAPSASQMFSFLRNSD
VPLEKNPKLKTHAMSVFVMTCEAAAQLRKAGKVTVRDTTLKRLGATHFKYGVGDAHFEVTRFALL
ETIKEAVPVDMWSPAMKSAWSEAYNQLVAAIKQEMKPAE

SEQ ID NO:15 *Arabidopsis thaliana*
MESEGKIVFTEEQEALVVKSWSVMKKNSAELGLKLFIKIFEIAPTTKKMFSFLRDSPIPAEQNPK
LKPHAMSVFVMCCESAVQLRKTGKVTVRETTLKRLGASHSKYGVVDEHFEVAKYALLETIKEAVP
EMWSPEMKVAWGQAYDHLVAAIKAEMNLSN

FIG. 1-CONT.

SEQ ID NO:16 *Pisum sativum*
MGFTDKQEALVNSSWESFKQNLSGNSILFYTIILEKAPAAKGLFSFLKDTAGVEDSPKLQAHAEQ
VFGLVRDSAAQLRTKGEVVLGNATLGAIHVQRGVTDPHFVVVKEALLQTIKKASGNNWSEELNTA
WEVAYDGLATAIKKAMT

SEQ ID NO:17 *Vigna unguiculata*
MVAFSDKQEALVNGAYEAFKANIPKYSVVFYTTILEKAPAAKNLFSFLANGVDATNPKLTGHAEK
LFGLVRDSAAQLRASGGVVADAALGAVHSQKAVNDAQFVVVKEALVKTLKEAVGDKWSDELGTAV
ELAYDELAAAIKKAY

SEQ ID NO:18 *Bos taurus*
MGLSDGEWQLVLNAWGKVEADVAGHGQEVLIRLFTGHPETLEKFDKFKHLKTEAEMKASEDLKKH
GNTVLTALGGILKKKGHHEAEVKHLAESHANKHKIPVKYLEFISDAIIHVLHAKHPSDFGADAQA
AMSKALELFRNDMAAQYKVLGFHG

SEQ ID NO:19 *Sus scrofa*
MGLSDGEWQLVLNVWGKVEADVAGHGQEVLIRLFKGHPETLEKFDKFKHLKSEDEMKASEDLKKH
GNTVLTALGGILKKKGHHEAELTPLAQSHATKHKIPVKYLEFISEAIIQVLQSKH
PGDFGADAQGAMSKALELFRNDMAAKYKELGFQG

SEQ ID NO:20 *Equus caballus*
MGLSDGEWQQVLNVWGKVEADIAGHGQEVLIRLFTGHPETLEKFDKFKHLKTEAEMKASEDLKKH
GTVVLTALGGILKKKGHHEAELKPLAQSHATKHKIPIKYLEFISDAIIHVLHSKH
PGDFGADAQGAMTKALELFRNDIAAKYKELGFQG

SEQ ID NO:21 *Nicotiana benthamiana*
MSSFTEEQEALVVKSWDSMKKNAGEWGLKLFLKIFEIAPSAKKLFSFLKDSNVPLEQNAKLKPHS
KSVFVMTCEAAVQLRKAGKVVVRDSTLKKLGATHFKYGVADEHFEVTKFALLETIKEAVPEMWSV
DMKNAWGEAFDQLVNAIKTEMK

SEQ ID NO:22 *Bacillus subtilis*
MGQSFNAPYEAIGEELLSQLVDTFYERVASHPLLKPIFPSDLTETARKQKQFLTQYLGGPPLYTE
EHGHPMLRARHLPFPITNERADAWLSCMKDAMDHVGLEGEIREFLFGRLELTARHMVNQTEAEDR
SS

SEQ ID NO:23 *Corynebacterium glutamicum*
MTTSENFYDSVGGEETFSLIVHRFYEQVPNDDILGPMYPPDDFEGAEQRLKMFLSQYWGGPKDYQ
EQRGHPRLRMRHVNYPIGVTAAERWLQLMSNALDGVDLTAEQREAIWEHMVRAADMLINSNPDPH
A

SEQ ID NO:24 *Synechocystis* PCC6803
MSTLYEKLGGTTAVDLAVDKFYERVLQDDRIKHFFADVDMAKQRAHQKAFLTYAFGGTDKYDGRY
MREAHKELVENHGLNGEHFDAVAEDLLATLKEMGVPEDLIAEVAAVAGAPAHKRDVLNQ

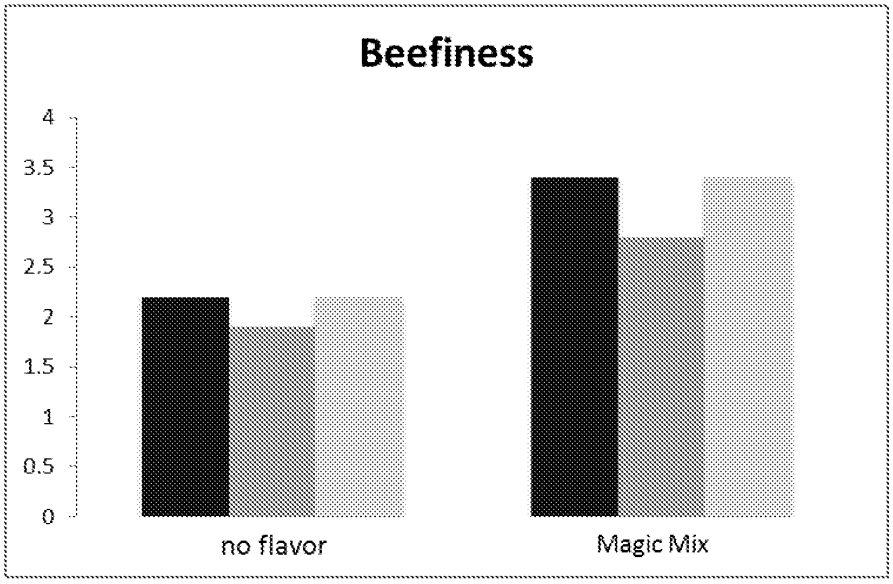
SEQ ID NO:25 *Synechococcus* sp. PCC 7335
MDVALLEKSFEQISPRAIEFSASFYQNLFHHHPELKPLFAETSQTIQEKKLIFSLAAIIENLRNP
DILQPALKSLGARHAEVGTIKSHYPLVGQALIETFAEYLAADWTEQLATAWVEAYDVIASTMIEG
ADNPAAYLEPELTFYEWLDLYGEESPKVRNAIATLTHFHYGEDPQDVQRDSRG

FIG. 1-CONT.

SEQ ID NO:26 *Nostoc commune*
MSTLYDNIGGQPAIEQVVDELHKRIATDSLLAPVFAGTDMVKQRNHLVAFLAQIFEGPKQYGGRP
MDKTHAGLNLQQPHFDAIAKHLGERMAVRGVSAENTKAALDRVTNMKGAILNK

SEQ ID NO:27 *Bacillus megaterium*
MREKIHSPYELLGGEHTISKLVDAFYTRVGQHPELAPIFPDNLTETARKQKQFLTQYLGGPSLYT
EEHGHPMLRARHLPFEITPSRAKAWLTCMHEAMDEINLEGPERDELYHRLILTAQHMINSPEQTD
EKGFSH

FIG. 2



Attorney Docket No.: 38767-0015001 / Client Ref: M3CON-Track 1

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DECLARATION (37 CFR 1.63) FOR UTILITY OR DESIGN APPLICATION USING AN APPLICATION DATA SHEET (37 CFR 1.76)

Title of Invention	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES

As the below named inventor, I hereby declare that:

This declaration is directed to:

[ ] The attached application, or

[X] United States application or PCT international application number 14/797,006

filed on July 10, 2015.

The above-identified application was made or authorized to be made by me.

I believe that I am the original inventor or an original joint inventor of a claimed invention in the application.

I hereby acknowledge that any willful false statement made in this declaration is punishable under 18 U.S.C. 1001 by fine or imprisonment of not more than five (5) years, or both.

WARNING:

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LEGAL NAME OF INVENTOR

Inventor: Rachel Fraser Date (Optional): 2/10/16

Signature: Rachel Fraser

Note: An application data sheet (PTO/SB/14 or equivalent), including naming the entire inventive entity, must accompany this form or must have been previously filed. Use an additional PTO/AIA/01 form for each additional inventor.

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The above-identified application was made or authorized to be made by me.

I believe that I am the original inventor or an original joint inventor of a claimed invention in the application.

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LEGAL NAME OF INVENTOR

Inventor: Patrick O'Reilly Brown M.D., Ph.D. Date (Optional) : _____

Signature: 

Note: An application data sheet (PTO/SB/14 or equivalent), including naming the entire inventive entity, must accompany this form or must have been previously filed. Use an additional PTO/AIA/01 form for each additional inventor.

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LEGAL NAME OF INVENTOR

Inventor: Jessica Karr Date (Optional): 9.21.15

Signature:

Note: An application data sheet (PTO/SB/14 or equivalent), including naming the entire inventive entity, must accompany this form or must have been previously filed. Use an additional PTO/AIA/01 form for each additional inventor.

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LEGAL NAME OF INVENTOR

Inventor: Celeste Holz-Schietinger Date (Optional) :

Signature:

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LEGAL NAME OF INVENTOR

Inventor: Elysia Cohn Date (Optional): 10/14/15

Signature:

Note: An application data sheet (PTO/SB/14 or equivalent), including naming the entire inventive entity, must accompany this form or must have been previously filed. Use an additional PTO/AIA/01 form for each additional inventor.

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TRANSMITTAL FOR POWER OF ATTORNEY TO ONE OR MORE REGISTERED PRACTITIONERS

Application Number	
Filing Date	
First Named Inventor	Rachel Fraser
Title	METHODS FOR GENERATING MEAT-FLAVORS AND AROMAS - TRACK 1 CON
Art Unit	
Examiner Name	
Attorney Docket Number	38767-0015008

SIGNATURE of Applicant or Patent Practitioner

Signature	/M. Angela Parsons/	Date (Optional)	/March 6, 2018/
Name	M. Angela Parsons	Registration Number	44,282
Title (if Applicant is a juristic entity)			
Applicant Name (if Applicant is a juristic entity)			

NOTE: This form must be signed in accordance with 37 CFR 1.33. See 37 CFR 1.4(d) for signature requirements and certifications. If more than one applicant, use multiple forms.

☐ *Total of _____ forms are submitted.

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POWER OF ATTORNEY BY APPLICANT

I hereby revoke all previous powers of attorney given in the application identified in either the attached transmittal letter or the boxes below.

Application Number	Filing Date
14/797,006	July 10, 2015

(Note: The boxes above may be left blank if information is provided on the Transmittal.)

☐ I hereby appoint the Patent Practitioner(s) associated with the following Customer Number as my/our attorney(s) or agent(s), and to transact all business in the United States Patent and Trademark Office connected therewith for the application referenced in the attached transmittal letter, which may be an application filed after the date of this power of attorney, or identified above:

OR

☒ I hereby appoint Practitioner(s) named in the attached list as my/our attorney(s) or agent(s), and to transact all business in the United States Patent and Trademark Office connected therewith for the patent application referenced in the attached transmittal letter, which may be an application filed after the date of this power of attorney, or identified above.

Please recognize or change the correspondence address for the application identified in the attached transmittal letter to:

☐ The address associated with the above-mentioned Customer Number

OR

☒ The address associated with Customer Number: **26191**

OR

Firm or Individual Name					
Address					
City		State		Zip	
Country					
Telephone		Email			

I am the Applicant (if the Applicant is a juristic entity, list the Applicant name in the box):

Impossible Foods Inc.

☐ Inventor or Joint Inventor (title not required below)

☐ Legal Representative of a Deceased or Legally Incapacitated Inventor (title not required below)

☒ Assignee or Person to Whom the Inventor is Under an Obligation to Assign (provide signer's title if applicant is a juristic entity)

☐ Person Who Otherwise Shows Sufficient Proprietary Interest (e.g., a petition under 37 CFR 1.46(b)(2) was granted in the application or is concurrently being filed with this document) (provide signer's title if applicant is a juristic entity)

SIGNATURE of Applicant for Patent

The undersigned (whose title is supplied below) is authorized to act on behalf of the applicant (e.g., where the applicant is a juristic entity).

Signature	Myra Pasek	Date (Optional)	Oct 17, 2016
Name	Myra Pasek		
Title	General Counsel		

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Attorney's Docket No.: 38767-0015001 / M3CON-Track 1

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

First Named Inventor	:	Rachel Fraser	Art Unit	:	1791
Serial No.	:	14/797,006	Examiner	:	Elizabeth A. Gwartney
Filed	:	July 10, 2015	Conf. No.	:	8496
Title	:	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES			

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 37 CFR §1.132

I, Celeste Holz-Schietinger, Ph.D., hereby declare as follows:

1. I am currently a Principal Scientist at Impossible Foods in Redwood City, California. I have held this position since 2015, and I am responsible for leading the Flavor Department. In this role, I direct flavor technology research as well as flavor product integration. I began working for Impossible Foods in 2012 as a Scientist, and I was elevated to a Senior Scientist in 2013. While at Impossible Foods, I have been the lead chemist on the identification, characterization and manipulation of biological flavor catalysis and potentiation, including through the use of heme-containing proteins.

2. I received my Bachelors of Science (B.S.) degree in Molecular Biology and Evolution, with a minor in Chemistry, in 2005 from The Evergreen State College in Olympia, Washington, and I received my Doctor of Philosophy (Ph.D.) degree in Biochemistry from the University of California at Santa Barbara in 2012. I was an Enzymology Research Associate at the University of California at Santa Barbara from 2007 to 2012, and I also was a Biochemistry Visiting Associate at Stanford University in 2009.

3. A copy of my curriculum vitae is attached as **Exhibit A**.

4. I am a named inventor on the above-referenced patent application.

5. I have read U.S. Application No. 14/797,006 ("the '006 application") as well as the Office Action mailed June 6, 2016, in which the claims were rejected as being anticipated by Proulx et al. (2006, *J. Agric. Food Chem.*, 54:1518-22) and obvious over Hseih et al. (US 2012/0093994) in view of Proulx et al.

6. Gas Chromatography-Mass Spectroscopy (GCMS) and Gas Chromatography-Olfactory (GCO) methods were used to evaluate the production of volatile compounds during cooking of three flavor reaction mixtures, and in particular, to evaluate the production of volatile compounds that are associated with meaty aroma. The first flavor reaction mixture contained 1% (10 mg/ml) Leghemoglobin (LegH-only); the second contained 50 mM Ribose with 50 mM Cysteine (Ribose + Cysteine); and the third contained 1% Leghemoglobin with 50 mM Ribose and 50 mM Cysteine (LegH + Ribose + Cysteine). All flavor reaction mixtures were adjusted to pH 6.5 using 50 mM of potassium phosphate buffer and 100 mM of sodium chloride buffer. Two mls of each flavor reaction mixture were prepared in 10 ml GCMS glass vials (referred to as "flavor reaction mixture samples" or "samples" hereinafter). Each sample was analyzed in duplicate.

7. An Agilent 7890A GC system coupled with a Leco TruTOF HT system was used along with a Gerstel MultiPurpose Sampler for auto-sampling. The Gerstel auto-sample was used to cook, cool, and perform HS-SPME on each of the samples. Each sample was heated to 150°C for 3 minutes with 750 rpm of agitation to "cook" the material. This was followed by a two-phase active cooling step that decreased the agitator temperature from 150°C to 50°C. The sample was extracted for 20 minutes at 50°C with 500 rpm of agitation using a 50/30 µm Divinylbenzene / Carboxen / Polydimethylsiloxane (DVB/CAR/PDMS), Stable flex, 23 Ga, Autosampler (Supleco, Cat# 57298-U) fiber.

8. Extracted samples were run on the GCMS by desorbing the SPME fiber in the Gerstel controlled CIS inlet for 6 minutes at 240°C. The desorbed sample was separated on a 60-meter wax column (Agilent, VF-Waxms 60 m x 0.25 mm x 0.25 mm, Part# CP9207) using a 35 minutes GCMS method (35°C for 2 minutes, ramp of 5C°/min until 172°C, ramp of 60C°/min until 255°C, 255°C for 4 minutes) and 1.75 mL/min Helium flow rate and splitless mode. Separated compounds were analyzed by the mass spectrometer, and all data was collected over a 14-500 µ mass range, with an acquisition rate of 10 spectra/sec, and a detector voltage off-set of 100.

9. Samples were then analyzed using Leco ChromaTOF optimized for TruTOF HT (Version 4.51.6.0) coupled with the NIST MS Search 2.2. The identity of each peak in each sample was identified in a two-step process. First, the mass spectrum of each peak with a signal

to noise greater than 50 was matched to a mass spectrum in the NIST library using a match similarity score of 650, which in my experience is a conservative match similarity score. Additionally, an internally developed calibration method was applied to the data set to confirm the identity of the compounds of interest. In the second step, the Statistical Compare function of ChromaTOF was used to align the named analytes across all samples in a set. The criteria for aligning a single peak across all samples were a similarity score (similarity between the peak spectra across all samples) of 700, a maximum retention time difference of 3 secs, and the peak must have been present in at least two samples.

10. The amount of each volatile compound was determined by averaging the signal intensities from the duplicate experiments. The amount of each volatile compound that can be attributed to Leghemoglobin (LegH)[1] was determined by using the average signal intensity for each volatile compound produced in the LegH-only sample as a baseline to standardize the average signal intensity for the same volatile compound produced in the LegH + Ribose + Cysteine sample. The fold-change in each volatile compound was determined by dividing the signal intensity produced in the presence of LegH (i.e., in the LegH + Ribose + Cysteine sample) by the signal intensity produced in the absence of LegH (i.e., the Ribose + Cysteine sample). If the signal intensity in the absence of LegH (i.e., the Ribose + Cysteine sample) was not detectable, the volatile compound produced in the presence of LegH (i.e., in the LegH + Ribose + Cysteine sample) was indicated as "novel," meaning that the volatile compound was newly produced (within the limits of detection of the technique) in the LegH + Ribose + Cysteine sample.

11. The results of the above-described experiments are shown in **Exhibit B**. The first column shows a list of volatile compounds, grouped into eight (8) different classes of chemicals. The next three columns show the amount of each volatile compound, calculated as described above, for each of the samples (LegH only, Ribose + Cysteine, and LegH + Ribose + Cysteine) followed by a column indicating the fold-change, calculated as described above, or indicating that the volatile compound is novel as described above. The next column provides, where

[1] Volatile compounds that can be attributed to LegH refer to compounds that are, independently, either not detectably produced or produced in a detectably smaller amount upon cooking compared with the control samples (e.g., LegH-alone or Ribose + Cysteine in the absence of LegH).

available, one or more peer-reviewed publications in which that particular volatile compound is associated with meaty aroma.

12. The presence of LegH resulted in the production of **twenty-eight (28)** novel volatile compounds that were not produced in the absence of LegH. The 28 novel volatile compounds can be divided into eight (8) different classes of compounds as follows. Representative volatile compounds in each class are shown below; the full list can be found in **Exhibit B**.

- *Oxygen (O)- and/or Nitrogen (N)-containing heterocyclic*: six (6) novel O- and/or N-containing heterocyclic compounds were produced (i.e., 2-methyl furan; 2-pentyl furan; bis(2-methyl-3-furyl) disulfide; 2,5-dimethyl-pyrazine; methyl-pyrazine; and 2-ethyl-5-methyl-pyrazine);
- *Sulfur (S)-containing heterocyclic*: nine (9) novel S-containing heterocyclic compounds were produced (e.g., 2-methyl-3-furanthiol; 3-thiophenemethanol; and 2-methyl-thiazole);
- *sulfides*: one (1) novel sulfide compound was produced (i.e., dimethyl sulfide);
- *aldehydes*: four (4) novel aldehydes were produced (e.g., (E,E)-2,4-hexadienal; and decanal);
- *lactones*: two (2) novel lactones were produced (i.e., 5-ethyldihydro-2(3H)-furanone; and dihydro-5-pentyl-2(3H)-furanone);
- *ketones*: three (3) novel ketones were produced (e.g., 2-octanone; and 3-nonanone);
- *alcohols*: one (1) novel alcohol was produced (i.e., p-cresol); and
- *carboxylic acids*: two (2) novel carboxylic acids were produced (i.e., hexanoic acid; and hexadecanoic acid).

Significantly, each of the 28 different novel volatile compounds have been associated, directly or indirectly, with a meaty aroma; see, for example, the references provided in **Exhibit B**. Of further significance, the presence of LegH in the flavor reaction mixture with Cysteine and Ribose resulted in the generation of two (2) novel volatile compounds upon heating: **2-methyl-3-furanthiol** and **bis(2-methyl-3-furyl)disulfide**. As noted in the Cadwallader

Declaration, which I have reviewed, both compounds are important aroma compounds in cooked meat and have extremely low odor detection thresholds of 0.7 parts-per-trillion (pptr) and 0.02 pptr, respectively (Belitz et al., 2006, Food Chemistry, Springer-Verlag, Berlin, p. 368). Both compounds usually are only present in low abundance (pptr range) in cooked meat (Tang et al., 2013, J. Sulfur Chem., 34:38-47). Despite their low abundance in meat, they are still among the most impactful aroma compounds because of their extremely low odor detection thresholds.

13. The presence of LegH in the flavor reaction mixture with Cysteine and Ribose also resulted in a synergistic and dramatic increase in the amount of **two (2)** volatile compounds produced upon heating compared to the amount of the same volatile compounds produced upon heating in the absence of LegH (i.e., in the Ribose + Cysteine sample) or in the LegH-only sample. In the presence of LegH, the amount of:

- 2-acetylthiazole was increased **2,851-fold**; and
- thiophene was increased **89-fold**.

Significantly, 2-acetylthiazole and thiophene have been associated with a meaty aroma. See, for example, the references cited in **Exhibit B**.

14. In sum, the presence of LegH with Cysteine and Ribose dramatically increased the number and amount of aroma compounds associated with meat upon cooking.

15. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

Respectfully submitted,

Date: Dec 2nd 2016

Celeste Holz-Schietinger, Ph.D.

EXHIBIT A

Celeste Holz-Schietinger, PhD

Principal Scientist, Impossible Foods

Redwood City, CA
celeste.holz@impossiblefoods.com

Innovative R&D Scientist and Product Developer responsible for leading the Flavor Department at Impossible Foods. Technical skills include enzymology, flavor chemistry, sensory science, lipid and protein biochemistry, protein purification, analytical chemistry, metabolic pathways and small molecule identification.

Education

Ph.D. Biochemistry, **University of California** at Santa Barbara *2007-2012*
from the Biomolecular Science and Engineering Program

B.S. Molecular Biology & Evolution, minor in Chemistry, *2001- 2005*
The Evergreen State College, Olympia, WA

Research

Principal Scientist, *Impossible Foods, Dr. Pat Brown* *2015- Current*

Senior Scientist, *Impossible Foods, Dr. Peg Scott, Dr. Chris Davis* *2013- 2015*

Scientist, *Impossible Foods, Dr. Chris Davis* *2012-2013*

- R&D scientist and flavor department leader at Impossible Foods, directing flavor technology research and flavor product integration.
- Manage a team of scientists to understand the fundamental biology and chemistry of flavor perception and flavor creation. We identify the catalysis and precursors that create aroma compounds, determine how aroma compounds create particular flavors profiles, and then how to recreate these flavors in products.
- Lead chemist on the identification, characterization and manipulation of biological flavor catalysis, including heme-proteins, microbes, and enzymes.
- Support off flavor identification and removal in products and feedstock with product developers, protein biochemist and process engineers.
- Flavor product integration, leading efforts for creating meat replicas from plants that taste and smell like animal based meats, working closely with manufacturing to scale the process, and marketing to meet consumers desires.
- Previous roles also included but not limited to sensory panel leader, lipid group leader, and product developer creating nut-base cheese (Kite Hill Cheese).

Enzymology Research Associate, *UC Santa Barbara, Dr. Norbert Reich* *2007 –2012*

- Researched the underlying chemical mechanisms that regulate how the genetic code is translated to creates phenotypic variation in human cells; particularly, understanding the developmental enzymes (DNMT3B and DNMT3A) and their modulators.
- Identified the mechanisms and structural changes that occurred with mutated forms of DNMT3A, highly associated with diseased states that change gene expression.

- Particular specialty was the development and execution of kinetic and biophysical assays to characterize the enzymology and regulation of the *de novo* DNA methyltransferase (DNMT3A).
- Mentored and lead 6 undergraduate researchers.

Biochemistry Visiting Associate, *Stanford University, Dr. Pat Brown (collaboration 2008-2010) 06-11/2009*

- Global identification of RNA associated with developmental enzymes (DNMT3B and DNMT3A) through cellular differentiation and correlated to gene expression and DNA methylation patterns. Developed RNA library construction protocols for next generation sequencing, and became skilled in microarray, hES cell culture, and fluorescence microscopy.

Scientist in Endocrinology, *U.S. Geological Survey 05-09/2006*

- Conducted research on the stress response of translocation on the desert tortoise, though ELISA-based assays, leading team of 5 interns.

Molecular Biology NSF Fellow, *University of Georgia, Dr. Travis Glenn 05-09/2005*

- Conducted independent research on pair bonding in alligators and determining mutation rates by carrying out ABI Prism sequencing for SNP and microsatellites, multiplexed PCR, and sequence analyzes for genotyping.

Genetics Research Assistant, *Evergreen State College, Dr. Erik Thusen 2004–2005*

- Independent research on the population genetics of moon jellies, gaining experience in genomic DNA purification, PCR for species identification, and phylogenetic analyzes.

Developmental Biology Research Assistant, *Evergreen State College, Dr. Donald Morisato 2003–2004*

- Investigated the molecular mechanisms of generating the anterior-posterior axis in *Drosophila*, by carrying out *lac z* staining, *in situ* hybridization, and genetic crosses.

Environmental Science Research Assistant, *Pacific Crest Community School, Dr. Tigerin Peare 2000–2001*

- Independent research on the *E. coli* levels in the Willamette River, Portland, OR. Data collected results in new city testing and regulations on the city water treatment facilities.

Teaching/Mentoring Experience

Research Mentor for Undergraduate Students, *UC Santa Barbara 2008-2011*

- Lead undergraduate students in research projects; taught techniques and guided experiments.

Teaching Assistant for epigenetics, developmental & biology laboratory, *UCSB, intermittently 2007-2010*

- Prepared lectures, gave lectures, and instructed laboratory sections.

Teaching Assistant, *Santa Barbara City College, 2006 - 2007*

- Molecular biology, botany and general biology laboratories.

Scientific Computing Specialist, *Evergreen State College 2003 –2006*

- Taught and helped students with science software - including biological modeling, statistics, phylogenetic, mathematical, and graphical programs.

EXHIBIT B

Compound	Signal intensity (ND = Not detected (signal to noise of 50, ~ signal of > 20,000))			Fold Increase or Novel ((LegH + Ribose + Cysteine) - (LegH))/ (Ribose + Cysteine)	References
	LegH	Ribose + Cysteine	LegH + Ribose + Cysteine		
O- and N-Heterocyclic compounds					
Furan, 2-methyl-	ND	ND	162,329	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com
Furan, 2-pentyl-	ND	ND	3,574,602	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Moon et al. (2006, Food Res. Int., 39:294-308); Brewer (2006, "The Chemistry of Beef Flavor -- Executive Summary," National Cattlemen's Beef Association, #12814); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
bis(2-methyl-3-furyl) disulfide*	ND	ND	1,215,514	Novel	Mottram (1998, Food Chem., 62:415-24); Macleod (1994, The flavour of beef in "The falvour of Meat and Meat Products," ed Shahidi); Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); www.thegoodscentscompany.com
Pyrazine, 2,5-dimethyl-	ND	ND	224,117	Novel	Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97); www.thegoodscentscompany.com
Pyrazine, methyl-	1049328	ND	12,559,346	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor -- Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Pyrazine, 2-ethyl-5-methyl-	40894	ND	87,684	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
S-Heterocyclic compounds					
2-Methyl-3-furanthiol	ND	ND	793,456	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Mottram (1998, Food Chem., 62:415-24); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com;
2-Acetylthiazole	ND	153,183	436,716,305	2851	Song et al. (2012, J. Chromatog. A, 1254:115-24); McGorrin et al. (2001, Special Publication - Royal Soc. Chem., 274:67-84); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); www.thegoodscentscompany.com;
Dihydro-3-(2H)-thiophenone	ND	ND	1,168,594	Novel	www.thegoodscentscompany.com;
Thiophene, 3-methyl-	ND	1,127,036	2,650,365	2	Shibamoto (1980, J. Agric. Food Chem., 28:237-43)

3-Thiophenemethanol	ND	ND	430,476	Novel	Chen et al. (2000, J. Agric. and Food Chem., 48:3512-6)
Thiophene, 2,3-dihydro-5-methyl-	ND	ND	11,799,570	Novel	Elmore et al. (1999, J. Agric. Food Chem., 47:1619-25)
Thiophene	ND	2,422,423	216,296,040	89	Shibamoto (1980, J. Agric. Food Chem., 28:237-43); Macleod (1994, The flavour of beef in "The falvour of Meat and Meat products" ed Shahidi); www.thegoodscentscompany.com
3(2H)-Thiophenone, dihydro-2-methyl-	ND	ND	204,409	Novel	www.thegoodscentscompany.com
5-Methyl-2-thiophenecarboxaldehyde	ND	ND	3,046,036	Novel	Shibamoto (1980, J. Agric. Food Chem., 28:237-43); www.thegoodscentscompany.com;
3-Methyl-2-thiophenecarboxaldehyde	ND	ND	657,464	Novel	Shibamoto (1980, J. Agric. Food Chem., 28:237-43); www.thegoodscentscompany.com
Ethanone, 1-(4,5-dihydro-2-thiazolyl)-	ND	ND	1,304,816	Novel	www.thegoodscentscompany.com;
Thiazole, 2-methyl-	ND	ND	18,835,504	Novel	Shibamoto (1980, J. Agric. Food Chem., 28:237-43)
Sulfides					
Dimethyl sulfide	ND	ND	1,601,935	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Grigorakis et al. (2003, Aquaculture, 225:109-19); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
Aldehydes					
2,4-Hexadienal, (E,E)-	ND	ND	9,495,304	Novel	Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Grigorakis et al. (2003, Aquaculture, 225:109-19); www.thegoodscentscompany.com;
Benzaldehyde, 4-ethyl-	ND	ND	24,320,418	Novel	Morita et al. (2003, J. Sci. Food Agric., 83:289-97); www.thegoodscentscompany.com
Decanal	ND	ND	26,163	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97); Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
Undecanal	ND	ND	813,152	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Lactones					
2(3H)-Furanone, 5-ethyldihydro-	ND	ND	72,377,175	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24)
2(3H)-Furanone, dihydro-5-pentyl-	ND	ND	31,360,192	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24)
Ketones					

2-Octanone	ND	ND	1,242,778	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Grigorakis et al. (2003, Aquaculture, 225:109-19)
3-Nonanone	ND	ND	650,546	Novel	Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
3,5-Octadien-2-one, (E,E)-	ND	ND	537,770	Novel	Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Grigorakis et al. (2003, Aquaculture, 225:109-19); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com;
Alcohols					
p-Cresol	ND	ND	536,008	Novel	Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences)
Carboxylic Acids					
Hexanoic acid	ND	ND	14,935	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Selli et al. (2009, Food Chem., 114:317-22)
Hexadecanoic acid	ND	ND	1,959,921	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); www.thegoodscentscompany.com;
* in addition to being categorized as an O- and N-containing heterocyclic, bis(2-methyl-3-furyl) disulfide also can be categorized as a sulfide and as a S-containing heterocyclic compound.					

EXHIBIT A

Celeste Holz-Schietinger, PhD

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Redwood City, CA
celeste.holz@impossiblefoods.com

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Publications

Holz-Schietinger, C., Matje, D.M., Reich, N.O. Mutations in DNA methyltransferase (DNMT3A) observed in Acute Myeloid Leukemia patients disrupt methylation of clustered CpG sites. *J. Biol. Chem, 287: 30941*-30951 (2012).

Holz-Schietinger, C., and Reich, N.O. RNA modulation of the human *de novo* methyltransferase 3A (DNMT3A). *Nucleic Acids Res.* 40: 8550-8557 (2012).

Holz-Schietinger, C., Matje, D.M., Flexer Harrison, M. and Reich, N.O. DNA oligomerization controls the mechanism of *de novo* DNA methylation. *J. Biol. Chem.* 286, 41479-41488 (2011).

Holz-Schietinger, C. & Reich, N.O. The inherent processivity of the human *de novo* methyltransferase 3A (DNMT3A) is enhanced by DNMT3L. *J. Biol. Chem.* 285, 29091–29100 (2010).

Holz-Schietinger, C., Matje, D.M. and Reich, N.O. *De novo* DNA methyltransferase DNMT3A: regulation of oligomeric state and mechanism of action in response to physiological pH changes. *Biochimica et Biophysica Acta (BBA). 1850:1131-1139 (2015).*

Purdy, M.M., **Holz-Schietinger, C.** & Reich, N.O. Identification of a second DNA binding site in human DNA methyltransferase 3A by substrate inhibition and domain deletion. *Arch. Biochem. Biophys.* 498, 13-22 (2010).

Lance, S., Tuberville, T., Dueck, L., **Holz-Schietinger, C.,** Trosclair, P., Elsey, R., Glenn, T. Multi-year multiple paternity and mate fidelity in the American alligator, *Alligator mississippiensis. Molecular Ecology.* 2009. 18, 4508-4520 (2009).

Awards/Fellowships

- 2010-2011 Philip & Aida Siff Fellowship for excellent undergraduate student mentoring
- 2010, 2011 Best graduate presentation for USCB BMSE and MCDB departments
- 2009 Honorable Mention NSF Graduate Research Fellowship
- 2007-2008 Amgen Fellowship
- 2005 NFS Undergraduate Research Fellowship
- 2005 Evergreen foundation grant (research funding)
- 2002-2004 Evergreen State College foundation scholarship for academic excellence
- 2001-2002 USAA All-American Scholar, Philip S. Wilder Award for Academic Distinction, National Deans List

Training Courses /Research Conferences

- 14th Weurman Flavour Research Symposium, (2014) Cambridge, UK
- FKS Flavor Reaction Course, (2014), University of Missouri St Louis
- American Oil Chemistry Society (AOCS) conference, Lecithin Functions in Technology and Nutrition Course, (2013) AOCS Montreal QC, Canada
- Enzymes, Coenzymes & Metabolic Pathways Gordon conference, (2011, 2010) Waterville Valley, NH

EXHIBIT B

Compound	Signal intensity (ND = Not detected (signal to noise of 50, ~ signal of > 20,000))			Fold Increase or Novel ((LegH + Ribose + Cysteine) - (LegH))/ (Ribose + Cysteine)	References
	LegH	Ribose + Cysteine	LegH + Ribose + Cysteine		
O- and N-Heterocyclic compounds					
Furan, 2-methyl-	ND	ND	162,329	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com
Furan, 2-pentyl-	ND	ND	3,574,602	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Moon et al. (2006, Food Res. Int., 39:294-308); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
bis(2-methyl-3-furyl) disulfide*	ND	ND	1,215,514	Novel	Mottram (1998, Food Chem., 62:415-24); Macleod (1994, The flavour of beef in "The falvour of Meat and Meat Products," ed Shahidi); Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); www.thegoodscentscompany.com
Pyrazine, 2,5-dimethyl-	ND	ND	224,117	Novel	Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97); www.thegoodscentscompany.com
Pyrazine, methyl-	1049328	ND	12,559,346	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Pyrazine, 2-ethyl-5-methyl-	40894	ND	87,684	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
S-Heterocyclic compounds					
2-Methyl-3-furanthiol	ND	ND	793,456	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Mottram (1998, Food Chem., 62:415-24); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com;
2-Acetylthiazole	ND	153,183	436,716,305	2851	Song et al. (2012, J. Chromatog. A, 1254:115-24); McGorrin et al. (2001, Special Publication - Royal Soc. Chem., 274:67-84); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); www.thegoodscentscompany.com;
Dihydro-3-(2H)-thiophenone	ND	ND	1,168,594	Novel	www.thegoodscentscompany.com;
Thiophene, 3-methyl-	ND	1,127,036	2,650,365	2	Shibamoto (1980, J. Agric. Food Chem., 28:237-43)

3-Thiophenemethanol	ND	ND	430,476	Novel	Chen et al. (2000, J. Agric. and Food Chem., 48:3512-6)
Thiophene, 2,3-dihydro-5-methyl-	ND	ND	11,799,570	Novel	Elmore et al. (1999, J. Agric. Food Chem., 47:1619-25)
Thiophene	ND	2,422,423	216,296,040	89	Shibamoto (1980, J. Agric. Food Chem., 28:237-43); Macleod (1994, The flavour of beef in "The falvour of Meat and Meat products" ed Shahidi); www.thegoodscentscompany.com
3(2H)-Thiophenone, dihydro-2-methyl-	ND	ND	204,409	Novel	www.thegoodscentscompany.com
5-Methyl-2-thiophenecarboxaldehyde	ND	ND	3,046,036	Novel	Shibamoto (1980, J. Agric. Food Chem., 28:237-43); www.thegoodscentscompany.com;
3-Methyl-2-thiophenecarboxaldehyde	ND	ND	657,464	Novel	Shibamoto (1980, J. Agric. Food Chem., 28:237-43); www.thegoodscentscompany.com
Ethanone, 1-(4,5-dihydro-2-thiazolyl)-	ND	ND	1,304,816	Novel	www.thegoodscentscompany.com;
Thiazole, 2-methyl-	ND	ND	18,835,504	Novel	Shibamoto (1980, J. Agric. Food Chem., 28:237-43)
Sulfides					
Dimethyl sulfide	ND	ND	1,601,935	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Grigorakis et al. (2003, Aquaculture, 225:109-19); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
Aldehydes					
2,4-Hexadienal, (E,E)-	ND	ND	9,495,304	Novel	Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Grigorakis et al. (2003, Aquaculture, 225:109-19); www.thegoodscentscompany.com;
Benzaldehyde, 4-ethyl-	ND	ND	24,320,418	Novel	Morita et al. (2003, J. Sci. Food Agric., 83:289-97); www.thegoodscentscompany.com
Decanal	ND	ND	26,163	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97); Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
Undecanal	ND	ND	813,152	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Lactones					
2(3H)-Furanone, 5-ethyldihydro-	ND	ND	72,377,175	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24)
2(3H)-Furanone, dihydro-5-pentyl-	ND	ND	31,360,192	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24)
Ketones					

2-Octanone	ND	ND	1,242,778	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Grigorakis et al. (2003, Aquaculture, 225:109-19)
3-Nonanone	ND	ND	650,546	Novel	Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
3,5-Octadien-2-one, (E,E)-	ND	ND	537,770	Novel	Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Grigorakis et al. (2003, Aquaculture, 225:109-19); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com;
Alcohols					
p-Cresol	ND	ND	536,008	Novel	Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences)
Carboxylic Acids					
Hexanoic acid	ND	ND	14,935	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Selli et al. (2009, Food Chem., 114:317-22)
Hexadecanoic acid	ND	ND	1,959,921	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); www.thegoodscentscompany.com;
* in addition to being categorized as an O- and N-containing heterocyclic, bis(2-methyl-3-furyl) disulfide also can be categorized as a sulfide and as a S-containing heterocyclic compound.					

Attorney's Docket No.: 38767-0015001 / M3CON-Track 1

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

First Named Inventor	:	Rachel Fraser	Art Unit	: 1791
Serial No.	:	14/797,006	Examiner	: Elizabeth A. Gwartney
Filed	:	July 10, 2015	Conf. No.	: 8496
Title	:	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES		

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 35 U.S.C. § 1.132

I, Keith R. Cadwallader, Ph.D., hereby declare as follows:

1. I am currently a Professor of Food Chemistry in the Department of Food Science and Human Nutrition at the University of Illinois in Urbana, Illinois. I have held this position since 2007. I have worked and taught in the fields of Food Science, Food Chemistry and Flavor Science since 1985. Prior to my current position, I held Assistant/Associate Professor positions in Food Chemistry at Louisiana State University and Mississippi State University and was an Associate Professor at the University of Illinois.

2. I received my B.S. degree in Food Science from the University of Georgia in 1985, my M.S. degree in Food Science from the University of Florida in 1987, and my Ph.D. degree in Food Science (minor in Chemistry) from the University of Florida in 1990.

3. A copy of my curriculum vitae is attached as **Exhibit A**.

4. My research includes: chemistry and analysis of food and beverage flavors, flavor (aroma) characterization and flavor quality determination, instrumental and sensory identification and quantification of character-impact flavor compounds in foods/beverages and agricultural/ industrial materials, process flavor development, recovery of flavor constituents

from food processing by-products, development of methods to stabilize labile flavors, occurrence and mechanisms of flavor formulation in foods and beverages, technology for production of food and beverage flavors, and physical chemistry of food/beverage matrix-flavor binding, flavor release and flavor-packaging interactions. I have taught courses relating to food analysis, flavor chemistry, food products analysis, applied food chemistry, flavor and food acceptance, food composition, and flavor/color of foods.

5. I am a nationally and internationally recognized expert and scholar in the field of flavor science. I have co-edited five books and published 57 refereed book chapters and 96 refereed journal articles in the area of food flavor chemistry and analysis. My research has been published in well-respected journals in the field of flavor chemistry, including the *Journal of Agricultural and Food Chemistry*, *Food Chemistry*, *Journal of Food Science*, *Analytical Chemistry*, *Journal of Sensory Studies*, *Journal of the American Oil Chemists Society*, *Cereal Chemistry*, *Journal of Dairy Science*, *Flavour and Fragrance Journal* and others. I have presented more than 50 invited symposium talks and over 115 scientific papers at national and international scientific meetings. I am co-inventor of an invention described in a U.S. patent application related to stabilized compositions and methods of manufacture (Publication No. US 2014/0275543 A1).

6. Through my research and teaching, I have made both fundamental and applied contributions to the current understanding of flavor chemistry and analysis. In 2016, I received the University of Illinois, College of Agriculture, Consumer and Environmental Sciences, *Senior Faculty Award for Excellence in Research.* I was awarded the *2007 Stephen S. Chang Award for Excellence in Lipid or Flavor Science* from the Institute of Food Technologists. In 2010, I was

elected a *Fellow in the Agricultural and Food Chemistry Division of the American Chemical Society* in recognition of my scientific contributions to the field of agricultural and food chemistry, specifically in the area of flavor science.

7. I am a professional member of the Institute of Food Technologists and the American Chemical Society (Agricultural and Food Chemistry Division). From 2003-2005, I was the Chair and Program Chair of the Agricultural and Food Chemistry Division of the American Chemical Society. I currently serve on the editorial board of the *Flavour and Fragrance Journal* and as an associate editor of the *Journal of Food Science*.

8. In the preparation of my declaration, I have reviewed the U.S. Application Serial No. 14/797,006 ("the '006 application"), as well as the Office Action mailed June 6, 2016, in which the claims of the '006 application were rejected as being anticipated by Proulx et al. (2006, J. Agric. Food Chem., 54:1518-22) ("Proulx") and as being obvious considering Hsieh et al. (US 2012/0093994) ("the '994 application") in view of Proulx. I have also considered additional materials listed in **Exhibit B**.

9. Flavor is the integrated response to the simultaneous perception (total or combined sensation) of the taste, aroma and somatosensory (nerve response) characteristics of substances naturally present in foods. Although the peripheral sensory organs for detection of taste and smell stimuli are quite distinct, their signals are integrated in the orbitofrontal and other areas of the cerebral cortex of the brain to generate the perception of "flavor" and to mediate food recognition and overall acceptance (Chaudhari and Roper, 2010). It is generally accepted by most researchers that olfaction (aroma) plays the dominant role in the flavor of food (Spence, 2015), and aroma has been regarded as the single most important factor contributing to the

characteristic flavor of meat (Shahidi et al., 1986).

10. Gas chromatography-mass spectrometry (GC-MS) has been the method of choice for the analysis of aroma components of meat for over five decades (Cadwallader and MacLeod, 1998). Before GC-MS can be conducted, the volatile components of the product must first be isolated from the sample and then transferred to the instrument for subsequent analysis. Headspace solid phase microextraction (HS-SPME) combined with GC-MS has been widely and successfully used for analysis of aroma components of meat (Rochat et al., 2007) and "meat aroma" reaction mixtures (Jublot et al., 2010). During analysis, the GC-MS system separates the individual volatile compounds in the mixture and then the mass-spectrometer detects and characterizes each component individually, enabling both qualitative (compound identification) and quantitative (compound concentration) analyses to be performed.

11. In my opinion, Applicants discovered a "ground beef-like food product" containing no animal products and including:

0.01%-5% by weight of a heme-containing protein comprising an amino acid sequence having at least 80% sequence identity to the polypeptide set forth in SEQ ID NO:4;

a sugar, sugar alcohol, sugar acid, or sugar derivative selected from glucose, ribose, fructose, lactose, xylose, arabinose, glucose-6-phosphate, maltose, and galactose, and mixtures of two or more thereof; and

at least 10 mM of a compound selected from cysteine, cystine, selenocysteine, thiamine, methionine, and mixtures of two or more thereof; and

at least 10% or more by weight one or more plant proteins,

which has the surprising capability of promoting, upon heating, the efficient conversion of

nonvolatile flavor precursors into desirable meaty aroma compounds. This is a significant discovery since a person having ordinary skill in the art would not have anticipated that the inclusion of a heme protein would have any beneficial effect on flavor. In fact, iron, and specifically heme iron, has been most often associated with causing deleterious off-odor consequences in foods and especially meat products (Calkins, 2007). As a flavor scientist who has worked in the field for over 30 years, I consider Applicants' discovery to be completely unexpected.

12. In forming my opinion, I reviewed and considered Applicants' experiments related to their discovery and demonstration that the addition of heme protein (in some cases as leghemoglobin or LegH) is required or essential for the development of meaty aroma in a "ground beef-like food product" comprising no animal products. These experiments are described in the '006 application. In addition, I reviewed the experiments in the Declaration Under 37 CFR §1.132 of Dr. Celeste Holz-Schietinger, Ph.D. ("Holz-Schietinger Declaration").

13. The Hotz-Schietinger Declaration describes the application of state-of-the-art instrumental aroma analysis methods, specifically the aforementioned HS-SPME combined with GC-MS, to monitor the effect of LegH addition on the formation of volatile compounds, specifically those with meaty aroma characteristics, during heating (i.e., cooking). The objective was to demonstrate that LegH was capable of promoting or catalyzing the formation of meaty aroma compounds via the Maillard reaction and other thermal reactions between ribose and cysteine. Three flavor reaction systems were compared, consisting of: 1) 1% (10 mg/mL) LegH alone (**LegH control**), 2) 50 mM ribose + 50 mM cysteine alone (**Rib + Cyst**) and 3) 1% (10 mg/mL) LegH + 50 mM ribose + 50 mM cysteine (**LegH + Rib + Cyst**). The above reaction

mixtures were diluted in a buffer (50 mM potassium phosphate and 100 mM sodium chloride) and adjusted to pH 6.5, which is near the physiological pH of ground beef. A 2 mL aliquot of each reaction mixture was transferred to sample vials (sealed) which were subsequently heated (150C for 3 min) and then analyzed by HS-SPME-GC-MS.

14. It is my opinion that the above experimental approach and subsequent data analyses techniques used by Dr Celeste Holz-Schietinger are appropriate and acceptable for the purpose of evaluating and comparing aroma compound formation in the three reaction systems under heating conditions. It is also my opinion that these conditions which would simulate the cooking of a "ground beef-like food product" containing no animal products as described in the '006 application, and that the data obtained from the experimental approach would be reasonably predictive of the expected aroma compound formation from a "ground beef-like food product" containing no animal products and including the flavor reaction system components LegH, ribose, and cysteine.

15. In my opinion, Applicants' experiments in the '006 application clearly demonstrate that inclusion of LegH in the reaction mixture has the surprising result of greatly enhancing the reaction of ribose and cysteine to form a variety of volatile compounds possessing meaty aromas. Moreover, it is my opinion that Dr. Celeste Holz-Schietinger's experiments confirm these surprising results. Among the 28 compounds identified in her experiments, two in particular are well-known to possess meaty sensory characteristics and to be important and potent odorants in cooked meat. Specifically, 2-methyl-3-furanthiol (MFT) and its dimerized form bis(2-methyl-3-furyl)disulfide (MFT-MFT) are important aroma compounds in cooked meat (Tang et al., 2013). Both compounds have extremely low odor detection thresholds of 0.7

parts-per-trillion (pptr) and 0.02 pptr, respectively (Belitz et al. 2006), and are usually only present in low abundance (pptr range) in cooked meat (Tang et al., 2013). Despite their low abundance in meat they are still among the most impactful aroma compounds because of their extremely low odor detection thresholds.

16. On the basis of my considerable knowledge and experience in the field of flavor science, it is my opinion that Applicants have indeed discovered and demonstrated the unique ability of heme-protein, including LegH, to serve as an efficient catalyst or promoter of the Maillard reaction and/or other relevant thermal reactions (e.g. thiamine degradation) which can form meaty aroma compounds in a "ground beef-like food product" containing no animal products.

17. It is also my opinion that a person having ordinary skill in the art would not have anticipated or expected this discovery based on the prior art cited. My opinion takes into consideration the Proulx et al. (2006) study, which did not disclose, mention or otherwise anticipate the potential for a heme-protein to influence or promote the formation of desirable meaty aroma compounds. Furthermore, Applicants' unique flavor precursor system, in which a heme-protein is intentionally added to act as an essential promotor of meat flavor generation during cooking, was not disclosed, mentioned or otherwise anticipated in the '994 application. My opinion also takes into consideration the common understanding of those in the field that the inclusion of iron, including heme iron, has been most often associated with causing deleterious off-odor consequences in foods and especially meat products.

18. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these

statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

Respectfully submitted,

Date: December 3, 2016

Keith Cadwallader

EXHIBIT A

Keith R. Cadwallader

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and Human Nutrition
University of Illinois at Urbana-Champaign
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Champaign, IL 61822

Campus:
102 ABL, MC-640

Education

University of Florida, Gainesville, FL	Fall, 1990	Ph.D. Food Science (Chem. minor)
University of Florida	Summer. 1987	M.S. Food Science
University of Georgia, Athens, GA	Spring, 1985	B.S. Food Science

Professional Experience

2007-present. Professor of Food Chemistry, Department of Food Science & Human Nutrition, University of Illinois. **Research Interests:** Chemistry and analysis of food flavor. Food flavor (aroma) characterization and flavor quality determination. Use of instrumental and sensory methods to identify and quantify character-impact flavor compounds in foods/agricultural/industrial materials. Development of process flavors. Recovery of flavor constituents from food processing by-products. Measurement and prediction of flavor diffusion, release and binding. Development of methods to stabilize labile flavors.
1999-2007. Associate Professor of Food Chemistry, University of Illinois.
1996-1999. Associate Professor of Food Science, Food Science & Technology, Mississippi State Univ.
1994-1996. Assistant Professor of Food Science, Mississippi State University
1991-1994. Assistant Professor of Food Science, Dept. Food Science, Louisiana State University.
1985-1990. Pre-doctoral Research Fellow, Dept. Food Science & Human Nutrition, Univ. Florida.

Current Appointment (UIUC)

Research – 60%; Teaching – 40%

Awards/Honors

- 2016 College of ACES Senior Faculty Award for Excellence in Research, University of Illinois,
- 2010 Elected Fellow, Agricultural and Food Chemistry Division of the American Chemical Society.
- Co-recipient, 2008 Donald A. Holt Achievement Award, Illinois Council on Food and Agricultural Research, presented to the Illinois Center for Soy Foods leadership team.
- 2007 Stephen S. Chang Award for Excellence in Lipid or Flavor Science, Institute of Food Technologists.
- Co-recipient, 2007 College of ACES Team Award for Excellence, University of Illinois, presented to the Illinois Center for Soy Foods leadership team
- Co-recipient, 2005 Bor S. Luh International Award, Institute of Food Technologists
- UIUC Incomplete list of teachers rated as excellent (Spr00, Fall00, Fall01, Spr03, Spr04, Spr05, Fall05; Spr06; Spr08; Spr10; Fall12; Fall14)
- 1996 Alpha Zeta Outstanding Teaching Award
- 1985-1990 FMC Corp. Research Fellowship Recipient
- 1984 U. Georgia Food Science Department Faculty Scholarship Recipient

Professional Memberships

Institute of Food Technologists & IFT, Chicago Section
American Chemical Society & ACS, Division of Agricultural and Food Chemistry

Teaching Interests and Experience

Teaching interests include food analysis, food chemistry, flavor chemistry and analysis, and sensory evaluation of foods. Courses taught are listed below.

University of Illinois

FSHN 418, *Food Analysis*: *2 h lecture: 6 h lab.* Principles and application of the chemical, physical, and instrumental methods used to determine the constituents of foods; special considerations applicable to the analysis of certain foods.

FSHN 519, Flavor Chemistry and Analysis: *2 h lecture: 6 h lab.* a graduate level course that involves the discussion/demonstration of the chemistry, manufacture/delivery and analysis (sensory, instrumental, and physical) of food flavor.

Mississippi State University

FST 4114/6114 *Analysis of Food Products*: *2 h. lecture; 6 h lab.* Principles, methods and techniques for quantitative, physical, chemical, and/or biological analysis of food products and ingredients.

FST 4241/6241 *Applied Food Chemistry*: *2 h lab.* Basic laboratory experiments to provide understanding of the function and interactions of chemical components of food.

FST 8163 *Flavor and Food Acceptance*: *2 h lecture; 1 h demonstration.* Theory and application of sensory evaluation and analytical chemistry techniques for the analysis of food flavor.

Louisiana State University

FDSC 4050 *Food Composition and Analysis*: *2 h. lecture; 6 h lab.* Principles, methods and techniques for quantitative, physical, chemical, and/or biological analysis of food products and ingredients.

FDSC 7040 *Flavor and Colors of Foods*: *2 h lecture; 3 lab.* Methods of chemical, physical, and instrumental analysis of food colors and flavors; natural and synthetic flavorings and colorants.

Research Publications

Books Edited (5)

Cadwallader, K.R. and Chang, S.K.C. (Eds.). 2010. *Chemistry, Texture and Flavor of Soy.* ACS Symposium Series 1059, American Chemical Society, Washington, DC.

Cadwallader, K.R., Drake, M.A. and McGorrin, R.J. (Eds.) 2007. *Flavor of Dairy Products.* ACS Symposium Series 971, American Chemical Society, Washington, DC.

Cadwallader, K.R. and Weenen, H. (Eds.). 2003. *Freshness and Shelf Life of Foods.* ACS Symposium Series 836, American Chemical Society, Washington, DC.

Rouseff, R.L. and Cadwallader, K.R. (Eds.). 2001. *Headspace Analysis of Foods and Flavor: Theory and Practice.* Kluwer Academic/Plennum Publishers, New York.

Shahidi, F. and Cadwallader, K.R. (Eds.). 1997. *Flavor and Lipid Chemistry of Seafoods.* ACS Symposium Series 674. American Chemical Society, Washington, DC.

Patent Applications (1)

Cadwallader, K.R. and Fang, M.-C. 2014. Stabilized compositions and methods of manufacture. US Patent Application US 2014/0275543 A1.

Book Chapters (57)

Cadwallader, K.R. 2016. Flavor challenges in functional beverages. Chapter 3. In *Handbook of Functional Beverages and Human Health*. Alasalvar, C. and Shahidi, F. (Eds.), CRC Press, pp. 27-34.

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Burnside, E. and Cadwallader, K.R. 2014. Aroma components of commercial and self-prepared rum ethers. In *Current Topics in Flavor Chemistry and Biology: Proceedings of the 9th Wartburg Symposium on Flavor Chemistry & Biology*, T. Hofmann, W. Krautwurst, D., and Schieberle, P. (Eds.), Eigenverlag Deutsche Forschungsanstalt für Lebensmittelchemie, Freising, Germany, pp. 244-249.

Moran, L., Yin, Y. Cadwallader, K.R. and Padua, G.W. 2014. Testing tools and physical, chemical, and microbiological characterization of microencapsulated systems. In *Microencapsulation in the Food Industry: A Practical Implementation Guide*. Gaonkar, A., Vasisht, N, Khare, A. and Sobel, R. (Eds.). Academic Press/Elsevier Inc., Oxford, UK, pp. 323-352.

Lorjaroenphon, Y., Hausch, B.J. and Cadwallader, K.R. 2014. Formation and stability of 2,3-dehydro-1,8-cineol in a model carbonated beverage system. In *Flavor Science: Proceedings from the XIII Weurman Flavour Research Symposium*, Elsevier, Inc. Oxford, UK.

Fang, M. and Cadwallader, K.R. 2012. Improved synthesis of deuterium-labeled alkyl pyrazines for use in stable isotope dilution analysis. In *Recent Advances in the Analysis of Foods and Flavor*. Mussinan, C. et al. (Eds.). ACS Symposium Series 1098, American Chemical Society, Washington, DC, pp. 57-69.

Lahne, J. and Cadwallader, K.R. 2012. Streamlined analysis of potent odorants in distilled alcoholic beverages. In *Chemistry and Flavor of Alcoholic Beverages.* Qian, M. et al., (Eds.). ACS Symposium Series 1104, American Chemical Society, Washington, DC, pp. 37-53.

Cadwallader, K.R., Potts, D., Brisske-BeVier, L. and Mirarefi, S. 2011. Contribution of volatile sulfur compounds to the characteristic aroma of roasted garlic. In *Volatile Sulfur Compounds in Food.* Qian, M.C., Fan, X. and Mahattanatawee, K. (Eds.). ACS Symposium Series 1068, American Chemical Society, Washington, DC, pp. 137-151.

Genthner, L.R. and Cadwallader, K.R. 2011. Structure-odor relationships of selected esters with (Z,Z)-3,6-nonadiene backbone structures. In *Advances and Challenges in Flavor Chemistry & Biology: Proceedings of the 9th Wartburg Symposium on Flavor Chemistry & Biology*, T. Hofmann, W. Meyerhof, P. Schieberle, Eds., Eigenverlag Deutsche Forschungsanstalt für Lebensmittelchemie, Garching, Germany, pp. 123-128.

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Other Publications (UIUC, 2000-present)

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Invited Talks

Cadwallader, K.R., Yin, Y., Hausch, B. and Chen, F. 2016. Assuring accuracy in the quantitation of the unstable odorant 2-acetyl-1-pyrroline in aromatic rice. ACS Symposium: *Flavor Stability: Chemical Changes in Flavor Molecules, Food-Flavor Matrix Interactions, Flavor Encapsulation.* 252nd Americal Chemical Society Meeting. Philadelphia, PA, August 21-25.

Cadwallader, K.R. 2016. Occurrence, formation and stabilization of the potent odorant 2-acetyl-1-pyrroline. Meeting of the Illinois Heartland Local Section of the American Chemical Society, Bloomington, IL, May 16, 2016.

Ickes, C. and Cadwallader, K.R. 2016. Characterizing aroma components of rum. ACS Symposium: *Flavor Chemistry of Alcoholic Beverages.* 251st American Chemical Society Meeting, San Diego, CA, March 13-17.

Cadwallader, K.R., Hausch, B. and Yin, Y. Quest for stability of 2-acetyl-1-pyrroline: The key odorant in aromatic rice and pandan. 17th Food Innovation Asia Conference 2015, Bangkok, Thailand, June 18-19.

Ickes, C. and Cadwallader, K.R. 2015. Understanding the effects of ethanol-flavor interactions on flavor perception in alcoholic beverages. ACS Symposium: *Challenges in Applied Flavor Sciences.* ACS National Meeting, Boston, MA, August 16-20, AGFD paper no. 297.

Pang, X., Wu, J. and Cadwallader, K.R. 2014. Identification of predominant contributors to off-odors formed in thermally processed melon juice. ACS Symposium: *Chemical Changes During Processing.* 248th American Chemical Society Meeting, San Francisco, CA, August 10-14. AGFD paper no. 177.

Cadwallader, K.R. and Lapsongphon, N. 2012. Identification of key odorants in fish sauce. US-Korea Conference (UKC) 2012, Los Angeles, August 8-11.

Fang, M. and Cadwallader, K.R. 2011. Convenient synthesis of stable deuterium labeled alkyl pyrazines for use in stable isotope dilution assays. ACS Symposium: *Recent Advances in Analysis of Food Flavors.* 242nd American Chemical Society Meeting, Denver, CO, August 28 – September 1, 2011, AGFD paper no. 11.

Hausch, B. and Cadwallader, K.R. 2010. Flavor chemistry of lemon-lime flavored carbonated beverages. ACS Symposium: *Symposium Honoring Prof. Terry E. Acree on the Occasion of His 70th Year.* 240th ACS National Meeting, Boston, MA, August 22-26, AGFD paper no. 40.

Lahne, J. and Cadwallader, K.R. 2010. Streamlined analysis of potent odorants in distilled alcoholic beverages. ACS Symposium: *Flavor Chemistry of Alcoholic Beverages.* 240th ACS National Meeting, Boston, MA, August 22-26, AGFD paper no. 106.

Cadwallader, K.R. and Kim, H. 2010. Flavor chemistry of fish sauce. ACS Symposium: *Fermented Foods.* 239th ACS National Meeting, San Francisco, CA, March 21-25, AGFD paper no. 112.

Cadwallader, K.R. 2010. Flavor chemistry of tree nuts. IFT Symposium: *Nuts: Phytochemicals, Health Effects and Flavor.* IFT Annual Meeting and Food Expo, Chicago, IL, July 17-20, paper no. 054-05.

Lozano, P.R. and Cadwallader, K.R. 2009. Effect of deamidation on binding of flavor compounds by dehydrated soy protein isolates. ACS Symposium: *Food Proteins and Food-based Protein Chemistry* 237th ACS National Meeting, Finley, J., organizer, Salt Lake City, UT, March 22-26.

Cadwallader, K.R. 2008. Sensory evaluation of aroma models for flavor characterization. *Symposium in Honor of Ronald Buttery G. Kenneth Recipient of the 2008 Spencer Award for Outstanding Achievement in Agricultural and Food Chemistry*, Kansas City Section, American Chemical Society, Kansas City, MO, Oct 27.

Cadwallader, K.R. 2008. AGFD and 100 years of chemical research on grains and legumes. ACS Symposium: *AGFD 100th Anniversary Symposium*, Brine, C. and Tunick, M.H., organizers, 236th ACS National Meeting, Philadelphia, PA, August 17-21.

Liu, Y and Cadwallader, K.R. 2008. Aroma components of "Carlos" muscadine (*Vitis rotundifolia* Michx) grape juice. ACS Symposium: Flavor and Health Benefits of Small Fruits, Rimando, A.M. and Qian, M.C., organizers, 236th ACS National Meeting, Philadelphia, PA, August 17-21.

Lozano, P.R. and Cadwallader, K.R. 2008. Flavor binding by soy proteins. ACS Symposium: Chemistry, Texture and Flavor of Soy, Cadwallader, K.R. and Chang, S. K. C. Chang, organizers, 236th ACS National Meeting, Philadelphia, PA, August 17-21.

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Cadwallader, K.R. Lozano, P.R. and Kim, H. 2007. Challenges in the isolation and identification of key aroma components of complex functional foods. ACS Symposium: *Food Bioactives and Nutraceuticals: Production, Chemistry, Analysis and Health Effects: Symposium Honoring Fereidoon Shahidi*, Rimando, A.M., organizer. 234th ACS National Meeting, Boston, MA, August 19-23, AGFD Paper 92.

Cook, S., Cadwallader, K.R. and Kim, H. 2007. Identification of key thermally-derived odorants in cooked chicken liver. ACS Symposium: *Thermal Generation of Flavors and Off-flavors,* Ho, C.-T., Raghavan, S., McGorrin, R.J., organizers. 234th ACS National Meeting, August 19-23. AGFD Paper 206.

Cadwallader, K.R., Lorjaroenphon, Y., Kim, H. and Lee, S-Y. 2007. Evaluation of key odorants in chipotle pepper by quantitative analysis, calculation of odor-activity values and omission studies. 8th Wartburg Symposium on Flavor Chemistry & Biology, Eisenach, Germany, February 28 – March 2.

Cadwallader, K.R. 2007. Protein-based functional beverages and flavoring considerations. In IFT Symposium: *Functional Beverages and Human Health.* Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 28 - August 1, Session-Paper 174-02.

Cadwallader, K.R. 2006. Chemical and sensory characterization of food aroma: An academic approach. Meeting of the Society of Flavor Chemists, Chicago, IL, October 19.

Cadwallader, K.R. 2006. Flavor chemistry of fish and shellfish. ACS Symposium: Molecular Cuisine. (Tunick, M.H., Risch, S.J. and Johnson, S.C., organizers). 232nd ACS National Meeting, San Francisco, CA, September 10-14, AGFD paper 61.

Cadwallader, K.R. and Zhou, Q. 2006. Characteristic aroma components of tree nuts. ACS Symposium: Nutrition and Sensory Quality of Nuts. (Pattee, H.E.; Shahidi, F.; Yu, L., organizers). 331st ACS National Meeting, Atlanta, GA, March 26 – March 30, AGFD paper 40.

Cadwallader, K.R. 2006. Modern techniques to characterize complex food flavor systems. International Symposium on *Functional Foods*, ATAM (Nutrition Society of Mexico) and Institute of Food Technologists, Mexico City, Mexico, August 17.

Rotsachakul, P., Cadwallader, K.R. and Chaiseri, S. 2005. Comparison of DHS and solvent extraction-SAFE for GCO-dilution analysis of key aroma components of Thai fried chili paste. ACS Symposium: *Aroma-active Compounds in Foods and Reaction Flavors: Formation, Measurement and Stability.* (Ho, C.-T., Weerasinghe, D.K. and Cadwallader, K.R., organizers). 330th ACS National Meeting, Washington, DC, August 28 – September 1, AGFD paper 9.

Cadwallader, K.R., Bordignon, J.R., Wang, P. and Sugiharto, R. 2005. Identification of odorants responsible for off-flavor of corn gluten meal. ACS Symposium: *Grain-based Products: Health and Flavor, and Safety Aspects* (Cadwallader, K.R., Schieberle, P. and Morello, M.J., organizers). 330th ACS National Meeting, Washington, DC, August 28 – September 1, AGFD paper 167.

Cadwallader, K.R., Drake, M.A., Carunchia-Whetstine, M.E. and Singh, T.J. 2005. Characterisation of Cheddar cheese flavour by sensory directed instrumental analysis and model studies. 11th Weurman Flavour Research Symposium, Roskilde, Denmark, June 21-24.

Gnadt, T.A., Jasso, L. and Cadwallader, K.R. 2005. Characteristic aroma components of chipotle peppers. ACS Symposium: *Chemistry and Flavor of Hispanic Foods* (de Mejia, E. and Tunick, M.H., organizers). 229th ACS National Meeting, San Diego, CA, March 13-17.

Lozano, P.R., Nicolalde, C.L. and Cadwallader, K.R. 2005. Identification of characteristic aroma components of mate (*Ilex paraguariensis*) tea. ACS Symposium: *Chemistry and Flavor of Hispanic Foods* (de Mejia, E. and Tunick, M.H., organizers). 229th ACS National Meeting, San Diego, CA, March 13-17.

Lecona-Uribe, S., Loarca-Piña, G., Arcila-Lozano, C.C. and Cadwallader, K.R. 2005. Chemical composition of essential oil from Mexican orégano. ACS Symposium: *Chemistry and Flavor of Hispanic Foods* (de Mejia, E. and Tunick, M.H., organizers). 229th ACS National Meeting, San Diego, CA, March 13-17.

Cadwallader, K.R. Importance of sample preparation technique on the composition of aroma extracts from fresh herbs. ACS Sympsosium: *Challenges in Herb Research – Chemical Analysis and Quality* (Ho, C.T. and Hwang, L.S., organizers). 227th ACS National Meeting, Anaheim, CA, March 28-April 1, 2004.

Cadwallader, K.R., Jarunrattanasri, A. and Theerakulkait, C. 2003. Aroma components of HVP made from rice bran protein. ACS Symposium: *Processed/Reaction Flavors, Ingredients and Intermediate Reactions.* (Weerasinghe, D.K. and Sucan, M.K., organizers). 226th ACS National Meeting, New York, NY, September 7-11.

Cadwallader, K.R. and Pojjanapimol, S. 2003. Characteristic aroma components of the cilantro mimics. ACS Symposium: *Natural Flavor and Fragrance Ingredients: Chemistry, Production and Application.* (Frey, C. and Rouseff, R.L., organizers). 226th ACS National Meeting, New York, NY, September 7-11.

Cadwallader, K.R. Relating Cheddar cheese flavor to chemical components. IFT Symposium: *Flavor Chemistry of Dairy Products.* (Drake, M.A., organizer). Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 12-16, #52-5.

Cadwallader, K.R., Pojjanapimol, S. and Chaiseri, S. 2002. Heat-induced changes in holy basil aroma components. ACS Symposium: *Challenges in the Characterization of Flavor Compound.* (Deibler, K.D. and Delwiche, J., organizers). 224th ACS National Meeting, Boston, MA, August 18-22.

Cadwallader, K.R., Zhou, Q. and Huda, M.S. 2002. Measuring flavor release from microencapsulants under controlled environmental conditions. ACS Symposium: *Controlled Release Systems for Foods.* (Shefer, S. and Yildiz, M.E., organizers). 223rd ACS National Meeting, Orlando, FL, April 7-11.

Cadwallader, K.R., Brehart, D.M., Schmidt, S.J. and Huda, M.S. 2002. Volatile release from a sucrose encapsulated flavoring. IFT Symposium: *Use of Carbohydrates for Encapsulation and Sustained Release.* (Obanni, M. and Asquith, T.N., organizers). Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #67-1.

Cadwallader, K.R., Lopez, J.R. and Menke, S.D. 2001. Aroma components of wines from Chardonel, a French-American hybrid grape variety. ACS Symposium: *Chemistry and Flavor of Nutritional Beverages.* (Shahidi, F. and Weerasinghe, D., organizers). 222nd ACS National Meeting, Chicago, IL, August 26-31.

Guthrie, B. and Cadwallader, K.R. 2001. Measuring flavor retention in dry food systems. IFT Symposium: *Developments in the Analysis of Complex Flavor Systems*. (Cadwallader, K.R. and McGorrin, R.J., organizers). Annual Meeting of the Institute of Food Technologies, New Orleans, LA, June 23-27, #76-2.

Cadwallader, K.R. 2001. Use of SPME Technology to Evaluate Antioxidant Effectiveness. Symposium: *Antioxidant Analysis*, (Engeseth, N.J., organizer). 92nd AOCS Annual Meeting and Expo, Minneapolis, MN, May 13-16.

Karagul-Yuceer,Y., Drake, M.A. and Cadwallader, K.R. 2001. Aroma characterization of fresh and stored nonfat dry milk. ACS Symposium: *Freshness and Shelf-life of Foods.* (Weenen, H. and Cadwallader, K.R., organizers). 222nd ACS National Meeting, Chicago, IL, August 26-30.

Cadwallader, K.R. 2000. Flavor Chemistry of Saffron. Symposium: *Carotenoid-Derived Aroma Compounds.* (Winterhalter, P. and Rouseff, R., organizers). 219th ACS National Meeting, San Francisco, CA, March 26-30, paper #46 AGFD.

Cadwallader, K.R. 1999. Sample dilution analysis for identification of aroma-impact components of edible oils. ACS Symposium: *Advances in Gas Chromatography/Olfactometry.* (Leland, J., Acree, T.E., organizers). 218th ACS National Meeting, New Orleans, LA, August 22-26.

Cadwallader, K.R. and Shahidi, F. 1999. Identification of potent odorants in seal blubber oil by direct thermal desorption-gas chromatography-olfactometry. ACS Symposium: *Chemistry and Nutrition of Highly Unsaturated Fatty Acids.* (Shahidi, F. and Finley, J.W., organizers). 217th ACS National Meeting, Anaheim, CA, March 21-25.

Cadwallader, K.R. and Howard, C.L. 1997. Analysis of aroma-active compounds in light-activated milk. ACS Symposium: *Challenges in the Isolation and Characterization of Flavor Compounds.* (Mussinan, C.J. and Morello, F., organizers). 214th National Meeting of the American Chemical Society, Las Vegas, NV, September 7-11.

Baek, H.-H. and Cadwallader, K.R. 1996. Character-impact aroma compounds of crustacea. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods.* (Shahidi, F. and Cadwallader, K.R., organizers). 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Cha, Y.J., Lee, G.H. and Cadwallader, K.R. 1996. Aroma-active compounds in salt-fermented anchovy. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods.* (Shahidi, F. and Cadwallader, K.R., organizers). 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Wang, H.-J., Cadwallader, K.R., Baek. H.H., D'Abramo, L.R. and Sullivan, J.A. 1996. Manipulating the flavor of crustacea using postharvest seawater acclimation. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods.* (Shahidi, F. and Cadwallader, K.R., organizers). 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Cadwallader, K.R. and Howard, C.L. 1996. Characterization of saffron flavor by aroma extract dilution analysis. ACS Symposium: *Spices: Flavor Chemistry and Antioxidant Properties.* (Risch, S.J. and Ho, C.T., organizers). 211th National Meeting of the American Chemical Society, New Orleans, LA, March 24-28.

Cadwallader, K.R., Baek, H.H., Chung, H.Y. and Moody, M.W. 1993. Contribution of lipid-derived components to the flavor of alligator meat. ACS Symposium: *Lipids and Food Flavors.* (Ho, C.T. and Hoffman, T.G., organizers). 205th National Meeting of the American Chemical Society, Denver, CO.

Braddock, R.J. and Cadwallader, K.R. 1993. Bioconversion of citrus d-limonene. Symposium paper presented at the 206th National Meeting of the American Chemical Society, Chicago, IL.

Braddock, R.J. and Cadwallader, K.R. 1991. Citrus by-products manufacture for food use. Symposium paper presented at the 52nd Annual Meeting of the Institute of Food Technologists, Dallas, TX, June 1-5.

Volunteered Abstracts

Kelley, L. and Cadwallader, K.R. 2016. Identification and quantitation of potent odorants in spearmint oils. 11th Wartburg Syposium on Flavor Chemistry & Biology, Eisenach, Germany, June 21-24.

McKenna, S. and Cadwallader, K.R. 2016. Identification and characterization of mint lactones: Trace-level odor-important compounds in peppermint oil. IFT Annual Meeting and Food Expo, Chicago, IL, July 17-19, paper no. PO2-020

Ickes, C., Lee, S. and Cadwallader, K.R. Novel creation of rum flavor lexicon through the use of web-based material. IFT Annual Meeting and Food Expo, Chicago, IL, July 17-19, paper no. P03-111

Yu, L., Thomas, L., Yin, L, Jerrell, J., Cadwallader, K. and Schmidt, S. 2015. Impact on sucrose crystal composition and chemistry on its thermal behavior. IFT Annual Meeting and Food Expo, Chicago, IL, July 11-14, paper no. 046-093.

Yu, L., Thomas, L., Yin, L, Jerrell, J., Cadwallader, K. and Schmidt, S. 2015. Impact on sucrose crystal composition and chemistry on its thermal behavior. IFT Annual Meeting and Food Expo, Chicago, IL, July 11-14, paper 046-093.

Cadwallader, K.R. and Fang, M.-C. 2014. A novel and attractive method for the stabilization of the potent odorant 2-acetyl-1-pyrroline. 14th Weurman Flavor Research Symposium, Cambridge, UK, September 15-19.

Donovan, J.D., Cadwallader, K.R. and Lee, Y. 2014. Improvement of protein microcapsule structure and core retention through the use of inulin. IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 181-01.

Erten, E. and Cadwallader, K.R. 2014. Characterization of aroma components of raw, dry roasted, and oil roasted almonds using solvent assisted flavor evaporation (SAFE) and aroma extract dilution analysis (AEDA). IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 206-26.

Lu, Y., Lee, J., Thomas, L., Jerrell, J., Cadwallader, K.R. and Schmidt, S.J. 2014. Investigating the thermal decomposition differences between beet and cane sugars using HPLC. IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 050-07.

Rosales, E., Cadwallader, K.R. and Engeseth, N. 2014. Effect of ultrasonic application on fatty acid composition and aroma compounds of dark chocolate. IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 206-94.

Burnsides, E. and Cadwallader, K. 2012 Comparison of aroma compounds in rums and rum ethers. IFT Annual Meeting and Food Expo, Las Vegas, NV, paper no. 193-28.

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Lee, H., Kim, H., Cadwallader, K.R. and Feng, H. 2011. Analysis of aroma compounds of apple cider treated by thermal-pasteurization, manosonication, thermosonication, and manothermosolication. IFT Annual Meeting and Food Expo, New Orleans, LA, June 11-14, paper no. 293-14.

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Andrews, L.S. and Cadwallader, K.R. 1993. Microbial and sensory quality of ground ginger as affected by gamma irradiation. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #817, p.206.

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Baek, H.H., Pike, M.M., Bidner, T.D., Moody, M.W., and Cadwallader, K.R. 1992. Evaluation of the off-odor associated with wild alligator. 53rd Annual Meeting of the Institute of Food Technologists, June 20-24, New Orleans, LA. Book of Programs and Abstracts, #359, p.93.

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Kim, H.R., Baek, H.H., Cadwallader, K.R., Meyers, S.P., and Godber, J.S. 1992. Improved flavor extractibility from crab waste using the hepatopancreatic enzyme of crawfish, *Procambarus clarkii*. 53rd Annual Meeting of the Institute of Food Technologists, June 20-24, New Orleans, LA, Book of Abstracts, #360, p.93.

Cadwallader, K.R. and Braddock, R.J. 1990. Enzymatic hydration of limonene to α-terpineol. 51st Annual Meeting of the Institute of Food Technologists, June 16-20, Anaheim, CA, Book of Programs and Abstracts, #354, p.170.

Cadwallader, K.R. 1988. Bioconversion of (+)-limonene. 49th Annual Meeting of the Institute of Food Technologists, June 19-22, New Orleans, LA, Book of Programs and Abstracts, #293.

Students/Postdoctoral/Visiting Scholar Researchers Supervised (1991-1999)

Visiting Scientists
Yong-Jun Cha (LSU, MSU) – Chang Won National University
Hetty Voermans (LSU) – University of Wegeningen, The Netherlands

Postdoctoral Researchers
Hyung-Hee Baek (MSU)

Graduate Students Completed (LSU/MSU)

MS Students
Sun-Kyun Yoo (LSU)
Min Cai (MSU)
Hsin-Jen (Connie) Weng (MSU)
Shi-Guei (Scott) Fu (MSU)
Shin-Pei (Peggy) Yang (MSU)
Yi-Fang (Grace) Wu (MSU)
Leyla Yuksel (MSU)
JiHyun Jung (MSU)
Min-Sheng Su (MSU)
Gwo-Yuh Li (MSU)
Jeong Heo (MSU)

Ph.D. Students
Hyung-Hee Baek (LSU)
Hau Yin Chung (LSU)
Gyu-Hee (Chang) Lee (MSU)
Ranee Surakarnkul (MSU)
Yonca Karagul-Yuceer (MSU)

Students/Postdoctoral/Visiting Scholar Researchers Supervised (UIUC, 2000-present)

Visiting Scholars
Byung Hak Ahn (2006-2007) - Korea Food Research Institute, South Korea
Supeeraya Arsa (2013-2014) – Kasetsart University, Bangkok, Thailand
Mantana Buttara (2011-present) – Suranaree Univeristy of Technology, Thailand
Dong Chen (2015-present) – China Agricultural University, Beijing, China

Rongrong Chen (2016-present) - Ever Maple Food Science and Technology Co., Ltd., China
Arporn Jarunrattanasri (2003-2004) – Naresuan University, Thailand
Young-Suk Kim (2015-present) – Ewha Womans University
Patthamawadi Kiatbenjaku. (2009-2011, 2012) – Suranaree University of Technology, Thailand
Fareeya Kulapichitr (2016-present) – Chulalongkorn University, Thailand
Sasinee Kunyaboon (2016-present) - Suranaree University of Technology, Thailand
Nawaporn Lapsongphon (2010-2011) - Suranaree University of Technology, Thailand
Yuping Liu (2014-2015) – Beijing Technology and Business University, China
Yaowapa Lorjaroenphone (2006-2007) – Kasetsart University, Thailand
Xueli Pang (2013-2014) – China Agricultural University, Beijing, China
Sompoche Pojjanapimol (2003-2004) - Kasetsart University, Thailand
Primseri Rotsachakul (2004-2005) – Kasetsart University, Thailand
Huanlu Song (2005-2006) – Beijing Technology and Business University, Beijing, China
Patcharaporn Tinchan (2010-2012) – Kasetsart University, Thailand
Jihong Wu (2011-2012) – China Agricultural University, Beijing, China

Postdoctoral Researchers (UIUC)
Masud Huda (2002-2003)
Orasa Suriyaphan (2001, 2003)
Tanoj Singh (2001-2005)
Hun Kim (2005-2012)

Graduate Students Completed (UIUC)
Daniel Benitez (MS 2003)
Laura Brisske (MS 2003)
Erin Burnside (MS 2012)
Gabriela Carrasco (MS 2007)
Shondra Cook (MS 2007)
Edibe Erten (Ph.D. 2016)
Ming Chih Fang (Ph.D.2013)
Samantha Gardiner (MS 2015)
Elizabeth Genthner (MS 2010; Ph.D. 2014)
Theresa Gnadt (MS 2007)
Julia Gregg (Nonthesis MS, CHEFS Program, 2009)
Bethany Hausch (MS 2010)
Lauren Kelley (MS)
Sean LaFond (MS 2009)
Jacob Lahne (MS 2010)
Yaowapa Lorjaroenphone (Ph.D. 2012)
Patricio Lozano (MS 2005 & Ph.D. 2009)
Samantha McKenna (MS, 2016)
Joshua Moore (MS 2006)
Linda Moran (MS 2013)
Ana Carolina Mosca (MS 2007)
Jason Niermann (Nonthesis MS, CHEFS Program 2001)
Sirima Puangpraphant (MS 2007)
Bethany Richardson (MS 2013)
Inthawoot Suppavorasatit (Ph.D. 2012)
Chenxing Sun (3+2 program, Nonthesis MS)
Qiaoxuan Zhou (MS 2001 & Ph.D. 2005)

Current Graduate Students (2016)
Courtney Beckett (MS)
Chelsea Ickes (Ph.D.)
Fengjia Chen (MS, 3+1)
Bethany Hausch (Ph.D.)
Nagiat Hwasa (Ph.D.)
Jing (Jenny) Jin (MS, 3+1)
Tiandan Wu (MS)
Wenqi Zhu (Ph.D.)
Yun Yin (Ph.D.)

Undergraduate Student Researchers
Kelsey Choquette (2012-2013)
Diniene Chreaton, Rap II (2001)
Ashley Davis, Rap II (ACES) (2001) & UIUC Ronald E. McNair Summer Research Program (2004)
Yaozheng (Tess) Liu (2013-present)
Rosemary Nash (2011-2012)
Mary Nowak (2004-2005)
Jieun Oh (2013)
Abigail Pishevar (2014-2015)
David Potts (2008-2010)
Stephanie Sajuti (2005-2007)
Dylan Troth (2015-present)
Katrina Vlahovich (2001-2002)

Involvement in Professional Organizations (2000-present)

Leadership

Chair and Program Chair for the Division of Agricultural and Food Chemistry (AGFD) of the American Chemical Society, 2003-2005. Responsibilities include organizing AGFD technical programs for two ACS National Meetings (fall 2004 and spring 2005 meetings).

Editorial Board

Flavour and Frangrance Journal

Associate Editor

Journal of Food Science – *Food Chemistry*

Symposia Organized

Cadwallader, K.R. and Shahidi, F. 2017. ACS Symposium: *Chemistry of Clean Ingredients.* Washington, DC, August 20-24.

Cadwallader, K.R. and Chang, S.K.C. 2008. ACS Symposium: *Chemistry, Texture and Favor of Soy,* Philadelphia, PA, August 17-21.

Cadwallader, K.R. 2008. ACS Symposium: *Recent Developments in Flavor Chemistry*. 235th ACS National Meeting, New Orleans, LA, April 6-10.

Ho, C.-T., Weerasinghe, D.K. and Cadwallader, K.R. 2005. ACS Symposium: *Aroma-active Compounds in Foods and Reaction Favors: formation, measurement and stability*. 330th ACS National Meeting, Washington, DC, August 28 – September 1.

Cadwallader, K.R., P. Schieberle and M.J. Morello. 2005. ACS Symposium: *Grain-based Products: Health and Flavor, and Safety Aspects*. 330th ACS National Meeting, Washington, DC, August 28 – September 1.

Cadwallader, K.R. *Obesity Management: Role of Soy Foods*. University of Illinois, Champaign, IL, May 25-26, 2005.

Cadwallader, K.R., Drake, M.A. and McGorrin, R.J. 2004. ACS Symposium: *Chemistry and Flavor of Dairy Products*. 228th ACS National Meeting, Philadelphia, PA, August 22-26.

Weenen, H. and Cadwallader, K.R. (organizers). 2001. ACS Symposium: *Freshness and Shelf-life of Foods*. 222nd ACS National Meeting, Chicago, IL, August 26-30.

Cadwallader, K.R. and McGorrin, R.J. (organizers). 2001. IFT Symposium: *Developments in the Analysis of Complex Flavor Systems*. Annual Meeting of the Institute of Food Technologies, New Orleans, LA, June 23-27.

Rouseff, R.L. and Cadwallader, K.R. 1998. ACS Symposium: *Headspace Analysis of Foods and Flavors*. 214th ACS National Meeting, Boston, MA, August 23-27.

Shahidi, F. and Cadwallader, K.R. 1996. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods*. 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Workshops/Short courses (organized)

A Place for Soy in a Better School Lunch Program. University of Illinois, Urbana, IL, March 12, 2003.

Traditional and Emerging Methods for the Manufacture and Analysis of Food Flavorings. Faculty of Agriculture Industry, King Mongkut's Institute of Technology Ladkraband (KMITL), Bangkok, Thailand, July 29, 2002

Soy Flavor Workshop. ICSF Workshop (Klein, B.P. et al. coorganizers). Illinois Center for Soy Foods, University of Illinois, Urbana, IL, April, 2002/2003.

Workshops/Short courses (lecturer)

Nutraceuticals, Ingredients, Challenges, Formulations, Flavours and Health Effects. International Society for Nutraceutical and Functional Foods. Denpasar, Bali, Indonesia, October 11 – 12.

Flavor Chemistry. (course co-taught with Drs. Chi-Tang Ho and Soo-Yeun Lee), Taiwan National University, May 10-19, 2008.

Flavor Research Workshop. ACS Short Course (Acree, T.E., course director), American Chemical Society, Boston, MA, August 17-18, 2007.

Flavor Research Workshop. ACS Short Course (Acree, T.E. and Deibler, K., organizers), American Chemical Society, August 25-26, 2005.

Cadwallader, K.R. Flavoring soy products: masking and creating. Preconference AOAC Short Course. *Soyfoods: Ingredients, Preparation, and Utilization,* Koseoglu, S. and Shahidi, F. (organizers), Cincinnati, OH, May 9, 2005.

Cadwallader, K.R. 2004. Aroma and flavor aspects of soy beverages. Preconference Shortcourse: *Functional Beverages,* Koseoglu, S and Shahidi, F. (organizers), Worldnutra: International Conference and Exhibition on Nutraceutacals and Functional Foods, San Francisco, CA, Nov 8, 2004

Flavor Research Workshop. ACS Short Course (Acree, T.E., organizer), American Chemical Society, Boston, MA, August 16-17, 2002; New York, NY, September 7-11, 2003.

Cheese Flavor and Texture: Sensory and Instrumental Analysis. SDFRC Short Course (Drake, M.A., course director), Southeast Dairy Foods Research Center, North Carolina State University, Raleigh, NC, August 7-9, 2002.

Proposal Reviewer/Panel Participation (2010-present)

Panelist/Reviewer (2015) - Office of Scientific Quality Review (OSQR)-USDA-ARS, NP 306 Panel 14: Functional Foods (2015)

Panelist/Reviewer (2015) - Office of Scientific Quality Review (OSQR)-USDA-ARS, NP 107 Panel 14: Grain Engineering (2014)

Panelist/Reviewer (2014) - Office of Scientific Quality Review (OSQR)-USDA-ARS, NP 107 Panel 5: Food Chemistry Analysis (2014)

Panelist/Reviewer (2013) – 2012/2013 King Abdulaziz City for Science and Technology (KACST) Research Proposal Competition

EXHIBIT B

MATERIALS CONSIDERED

- U.S. Patent Application No. 14/797,006 ("the '006 application")
- Office Action mailed June 6, 2016
- Proulx et al. (2006, J. Agric. Food Chem., 54:1518-22) ("Proulx")
- Hsieh et al. (US 2012/0093994) ("the '994 application")
- Declaration under 37 CFR § 1.132 of Dr. Celeste Holz-Schietinger ("the Holz-Schietinger Declaration")
- Lane and Nursten, "Variety of odors produced in Maillard model systems and how they are influenced by reaction conditions" in Maillard Reaction in Foods and Nutrition, Waller and Feather, eds., ACS Symposium Series No. 215, pp 141-158
- Kokkinidou and Peterson, 2013, Food Funct., 4:1093-104
- Elmore et al., 2001, "A comparison of headspace entrainment on Tenax with solid phase micro-extraction for the analysis of the aroma volatiles of cooked beef," in Headspace Analysis of Foods and Flavors; Rouseff and Cadwallader, eds, Springer
- Elmore et al. (2002, J. Agric. Food Chem., 50:1126-32)
- Chen et al. (2003, "Influence of DNA on Volatile Generation from Maillard Reaction of Cysteine and Ribose," *in* Nutraceutical Beverages, ACS Symposium Series Vol 871)
- Mottram and Nobrega (2002, J. Agric. Food Chem., 50(14):4080–86)
- Schieberle and Hofmann ("Characterization of Key Odorants in Dry-Heated Cysteine—Carbohydrate Mixtures: Comparison with Aqueous Reaction Systems," *in* Flavor Analysis, ACS Symposium Series, Vol 705)

- Mottram and Mottram (2002, "An Overview of the Contribution of Sulfur-Containing Compounds to the Aroma in Heated Foods," *in* Heteroatomic Aroma Compounds, ACS Symposium Series, Vol 826)
- Shu-C and Ho (1989, "Parameter Effects of the thermal reaction of cysteine and 2,5-dimethyl-4-hydroxy-3(2H)-furanone," *in* Thermal Generation of Aromas, ACS Symposium Series, Vol 409)
- Chemistry and Technology of Flavours and Fragrances, 2005, Ed David Rowe, Blackwell Publishing Ltd
- Flavor of Meat and Meat Products, 1994, Ed Shahidi, Springer-Science+Business Media, BV
- Handbook of Meat and Meat Processing, 2012, 2nd Ed, Ed Hui, CRC Press
- Chen and Ho (2002, J. Agric. Food Chem., 50(8):2372–76)
- Kerscher and Grosch (1998, J. Agric. Food Chem., 46:1954-58)
- Tang et al. (2013, J. Sulfur Chem., 34:38-47)
- Withcombe and Mussinan (1988, J. Food Sci., 53:658-60)

EXHIBIT A

Keith R. Cadwallader

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Home:
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Champaign, IL 61822

Campus:
102 ABL, MC-640

Education

University of Florida, Gainesville, FL	Fall, 1990	Ph.D. Food Science (Chem. minor)
University of Florida	Summer. 1987	M.S. Food Science
University of Georgia, Athens, GA	Spring, 1985	B.S. Food Science

Professional Experience

2007-present. Professor of Food Chemistry, Department of Food Science & Human Nutrition, University of Illinois. **Research Interests:** Chemistry and analysis of food flavor. Food flavor (aroma) characterization and flavor quality determination. Use of instrumental and sensory methods to identify and quantify character-impact flavor compounds in foods/agricultural/industrial materials. Development of process flavors. Recovery of flavor constituents from food processing by-products. Measurement and prediction of flavor diffusion, release and binding. Development of methods to stabilize labile flavors.
1999-2007. Associate Professor of Food Chemistry, University of Illinois.
1996-1999. Associate Professor of Food Science, Food Science & Technology, Mississippi State Univ.
1994-1996. Assistant Professor of Food Science, Mississippi State University
1991-1994. Assistant Professor of Food Science, Dept. Food Science, Louisiana State University.
1985-1990. Pre-doctoral Research Fellow, Dept. Food Science & Human Nutrition, Univ. Florida.

Current Appointment (UIUC)

Research – 60%; Teaching – 40%

Awards/Honors

- 2016 College of ACES Senior Faculty Award for Excellence in Research, University of Illinois,
- 2010 Elected Fellow, Agricultural and Food Chemistry Division of the American Chemical Society.
- Co-recipient, 2008 Donald A. Holt Achievement Award, Illinois Council on Food and Agricultural Research, presented to the Illinois Center for Soy Foods leadership team.
- 2007 Stephen S. Chang Award for Excellence in Lipid or Flavor Science, Institute of Food Technologists.
- Co-recipient, 2007 College of ACES Team Award for Excellence, University of Illinois, presented to the Illinois Center for Soy Foods leadership team
- Co-recipient, 2005 Bor S. Luh International Award, Institute of Food Technologists
- UIUC Incomplete list of teachers rated as excellent (Spr00, Fall00, Fall01, Spr03, Spr04, Spr05, Fall05; Spr06; Spr08; Spr10; Fall12; Fall14)
- 1996 Alpha Zeta Outstanding Teaching Award
- 1985-1990 FMC Corp. Research Fellowship Recipient
- 1984 U. Georgia Food Science Department Faculty Scholarship Recipient

Professional Memberships

Institute of Food Technologists & IFT, Chicago Section
American Chemical Society & ACS, Division of Agricultural and Food Chemistry

Teaching Interests and Experience

Teaching interests include food analysis, food chemistry, flavor chemistry and analysis, and sensory evaluation of foods. Courses taught are listed below.

University of Illinois

FSHN 418, *Food Analysis*: *2 h lecture: 6 h lab.* Principles and application of the chemical, physical, and instrumental methods used to determine the constituents of foods; special considerations applicable to the analysis of certain foods.

FSHN 519, Flavor Chemistry and Analysis: *2 h lecture: 6 h lab.* a graduate level course that involves the discussion/demonstration of the chemistry, manufacture/delivery and analysis (sensory, instrumental, and physical) of food flavor.

Mississippi State University

FST 4114/6114 *Analysis of Food Products*: *2 h. lecture; 6 h lab.* Principles, methods and techniques for quantitative, physical, chemical, and/or biological analysis of food products and ingredients.

FST 4241/6241 *Applied Food Chemistry*: *2 h lab.* Basic laboratory experiments to provide understanding of the function and interactions of chemical components of food.

FST 8163 *Flavor and Food Acceptance*: *2 h lecture; 1 h demonstration.* Theory and application of sensory evaluation and analytical chemistry techniques for the analysis of food flavor.

Louisiana State University

FDSC 4050 *Food Composition and Analysis*: *2 h. lecture; 6 h lab.* Principles, methods and techniques for quantitative, physical, chemical, and/or biological analysis of food products and ingredients.

FDSC 7040 *Flavor and Colors of Foods*: *2 h lecture; 3 lab.* Methods of chemical, physical, and instrumental analysis of food colors and flavors; natural and synthetic flavorings and colorants.

Research Publications

Books Edited (5)

Cadwallader, K.R. and Chang, S.K.C. (Eds.). 2010. *Chemistry, Texture and Flavor of Soy.* ACS Symposium Series 1059, American Chemical Society, Washington, DC.

Cadwallader, K.R., Drake, M.A. and McGorrin, R.J. (Eds.) 2007. *Flavor of Dairy Products.* ACS Symposium Series 971, American Chemical Society, Washington, DC.

Cadwallader, K.R. and Weenen, H. (Eds.). 2003. *Freshness and Shelf Life of Foods.* ACS Symposium Series 836, American Chemical Society, Washington, DC.

Rouseff, R.L. and Cadwallader, K.R. (Eds.). 2001. *Headspace Analysis of Foods and Flavor: Theory and Practice.* Kluwer Academic/Plennum Publishers, New York.

Shahidi, F. and Cadwallader, K.R. (Eds.). 1997. *Flavor and Lipid Chemistry of Seafoods.* ACS Symposium Series 674. American Chemical Society, Washington, DC.

Patent Applications (1)

Cadwallader, K.R. and Fang, M.-C. 2014. Stabilized compositions and methods of manufacture. US Patent Application US 2014/0275543 A1.

Book Chapters (57)

Cadwallader, K.R. 2016. Flavor challenges in functional beverages. Chapter 3. In *Handbook of Functional Beverages and Human Health*. Alasalvar, C. and Shahidi, F. (Eds.), CRC Press, pp. 27-34.

Yin, Y., Cadwallader, K.R. and Fang, M.-C. 2015. A novel method for the stabilization of the potent odorant 2-acety-1-pyrroline. In *Flavour Science: Proceedings of the XIV Weurman Flavour Research Symposium*, Taylor, A.J. and Mottram, D.S (Eds.), Context Products Ltd, Leicestershire, UK, pp. 571-574.

Burnside, E. and Cadwallader, K.R. 2014. Aroma components of commercial and self-prepared rum ethers. In *Current Topics in Flavor Chemistry and Biology: Proceedings of the 9th Wartburg Symposium on Flavor Chemistry & Biology*, T. Hofmann, W. Krautwurst, D., and Schieberle, P. (Eds.), Eigenverlag Deutsche Forschungsanstalt für Lebensmittelchemie, Freising, Germany, pp. 244-249.

Moran, L., Yin, Y. Cadwallader, K.R. and Padua, G.W. 2014. Testing tools and physical, chemical, and microbiological characterization of microencapsulated systems. In *Microencapsulation in the Food Industry: A Practical Implementation Guide*. Gaonkar, A., Vasisht, N, Khare, A. and Sobel, R. (Eds.). Academic Press/Elsevier Inc., Oxford, UK, pp. 323-352.

Lorjaroenphon, Y., Hausch, B.J. and Cadwallader, K.R. 2014. Formation and stability of 2,3-dehydro-1,8-cineol in a model carbonated beverage system. In *Flavor Science: Proceedings from the XIII Weurman Flavour Research Symposium*, Elsevier, Inc. Oxford, UK.

Fang, M. and Cadwallader, K.R. 2012. Improved synthesis of deuterium-labeled alkyl pyrazines for use in stable isotope dilution analysis. In *Recent Advances in the Analysis of Foods and Flavor*. Mussinan, C. et al. (Eds.). ACS Symposium Series 1098, American Chemical Society, Washington, DC, pp. 57-69.

Lahne, J. and Cadwallader, K.R. 2012. Streamlined analysis of potent odorants in distilled alcoholic beverages. In *Chemistry and Flavor of Alcoholic Beverages.* Qian, M. et al., (Eds.). ACS Symposium Series 1104, American Chemical Society, Washington, DC, pp. 37-53.

Cadwallader, K.R., Potts, D., Brisske-BeVier, L. and Mirarefi, S. 2011. Contribution of volatile sulfur compounds to the characteristic aroma of roasted garlic. In *Volatile Sulfur Compounds in Food.* Qian, M.C., Fan, X. and Mahattanatawee, K. (Eds.). ACS Symposium Series 1068, American Chemical Society, Washington, DC, pp. 137-151.

Genthner, L.R. and Cadwallader, K.R. 2011. Structure-odor relationships of selected esters with (Z,Z)-3,6-nonadiene backbone structures. In *Advances and Challenges in Flavor Chemistry & Biology: Proceedings of the 9th Wartburg Symposium on Flavor Chemistry & Biology*, T. Hofmann, W. Meyerhof, P. Schieberle, Eds., Eigenverlag Deutsche Forschungsanstalt für Lebensmittelchemie, Garching, Germany, pp. 123-128.

Kim, H.K. and Cadwallader, K.R. 2011. Instrumental analysis of seafood flavor. In *Handbook of Seafood Quality, Safety and Health Applications.* Alasalvar, C., Shahidi, F., Miyashita, K. and Wanasundara, U. (Eds.). Blackwell Publishing Ltd., UK, pp. 50-67.

Cadwallader, K.R., Moore, J.J., Zhang, Z. and Schmidt, S.J. 2010. Comparison of spray drying and Refractance Window™ drying technologies for the encapsulation of orange oil. In *Recent Advances in Food and Flavor Chemistry: Food Flavors and Encapsulation, Health Benefits, Analytical Methods, and Molecular Biology of Functional Foods.* Ho C.-T., Mussinan, C.J., Shahidi, F. and Tratras Contis, E. (Eds.). The Royal Society of Chemistry, Cambridge, UK, pp. 246-254.

Sun, C., Cadwallader, K.R. and Kim, H. 2010. Comparison of key aroma components between soymilks prepared by cold and hot grinding methods. In *Chemistry, Texture and Flavor of Soy.* Cadwallader, K.R. and Chang, S.K.C. (Eds.). ACS Symposium Series 1059, American Chemical Society, Washington, DC, pp. 361-373.

Suppavorasatit, I. and Cadwallader, K.R. 2010. Flavor-soy protein interactions. In *Chemistry, Texture and Flavor of Soy.* Cadwallader, K.R. and Chang, S.K.C. (Eds.). ACS Symposium Series 1059, American Chemical Society, Washington, DC, pp. 339-359.

Cadwallader, K.R., Tamamoto, L.C. and Sajuti, S.C. 2010. Aroma components of fresh and stored pomegranate (*Punica granatum* L.) juice. In *Flavors in Noncarbonated Beverages.* Da Costa, N.C. and Cannon, R.J. (Eds.). ACS Symposium Series 1036. American Chemical Society, Washington, DC, pp. 93-101.

Liu, G.-L. and Cadwallader, K.R. 2010. Aroma-impact components of "Carlos" muscadine grape juice. In *Flavor and Health Benefits of Small Fruits.* Qian, M.C. and Rimando, A.M. (Eds.). ACS Symposium Series 1035, American Chemical Society, Washington, DC, pp. 63-77.

Cadwallader, K.R. and Singh, T. 2009. Flavours and off-flavours in milk and dairy products. In *Advanced Dairy Chemistry-Volume 3. Lactose, Water, Salts and Minor Constituents.* McSweeny, P.L.H. and Fox, P.F. (Eds). Springer Science+Business Media, LLC, New York, pp. 631-690.

Cadwallader, K.R. and Puangpraphant, S. 2008. Volatile compounds in tree nuts. In Tree Nut s: Composition, Phytochemicals, and Health Effects. Alasalvar, C. and Shahidi, F. (Eds.) Taylor and Francis, LLC, Boca Raton, FL, pp. 109-125.

Singh, T. and Cadwallader, K.R. 2008. Cheese. In *Dairy Processing and Quality Assurance.* Chandan, R.C., Kilara, A. and Shah, N. (Eds.), Wiley-Blackwell, Danvers, MA, pp. 273-308.

Lorjaroenphon, Y., Cadwallader, K.R., Kim, H. and Lee, S-Y. 2008. Evaluation of key odorants in chipotle pepper by quantitative analysis, calculation of odor activity values and omission studies. In *Recent Highlights in Flavor Chemistry and Biology: Proceedings of the 8th Warburg Symposium on Flavor Chemistry & Biology,* Hofmann, T., Meyerhof, W. and Schieberle, P. (Eds.). Deutche Forschungsanstalt für Lebensmittelchemie, Garching, Germany, pp. 191-200.

Zhou, Q. and Cadwallader, K.R. 2008. Measurement of flavor-soy protein interactions in low-moisture solid food systems by inverse gas chromatography. Ch. 4. In *Food Flavor: Chemistry, Sensory Evaluation and Biological Activity.* Tamura, H., Ebeler, S.E., Kubota, K. and Takeoka, G.R. (Eds.). ACS Symposium Series 988. American Chemical Society, Washington, DC., pp. 45-54.

Cadwallader, K.R. 2007. Wood smoke flavor. In *Handbook of Meat, Poultry & Seafood Quality.* Nollet, L.M.L. (Ed.) and Boylsten, T., Chen, F. Coggins, P.C., Gloria, M.B., Hyldig, G., Kerth, C.R., McKee, L.H., Hui, Y.H. (Assoc. Eds.), Blackwell Publishing, Inc., Lancaster, PA, pp. 201-210.

Cadwallader, K.R. 2007. Measuring cheese flavour. In *Improving the Flavour of Cheese.* Weimer, B. (Ed.), Woodhead Publishing Ltd., Cambridge, UK, pp. 401-412.

Cadwallader, K.R., Singh, T.K. and Jerrell, J.P. 2007. Streamlined analysis of short-, medium- and long-chain free fatty acids in Dairy Products. In *Flavor of Dairy Products.* Cadwallader, K.R., Drake, M.A. and McGorrin, R.J. (Eds.). ACS Symposium Series 971, American Chemical Society, Washington, DC. pp. 111-122.

Drake, M.A., Cadwallader, K.R., Carunchia Whetstine, M.E. 2007. Establishing links between sensory and instrumental analyses of dairy flavors: Example Cheddar cheese. In *Flavor of Dairy Products.* Cadwallader, K.R., Drake, M.A. and McGorrin, R.J. (Eds.). ACS Symposium Series, American Chemical Society, Washington, DC., pp. 51-77.

Singh, T.K., Cadwallader, K.R. and Drake, M.A. 2007. Flavor of dairy products. In *Handbook of Food Products Manufacturing: Health, Meat, Milk, Poultry, Seafood, and Vegetables.* Hui, Y.H. (Ed.), John Wiley & Sons, Inc., Hoboken, NJ, pp. 715-748.

Drake, M.A., Miracle, R.E., Caudle, A.D. and Cadwallader, K.R. 2007. Relating sensory and instrumental analyses. In *Sensory-Directed Flavor Analysis.* Marsili, R. (Ed.), CRC Press/Taylor & Francis Group, LLC, Boca Raton, FL, pp. 23-54.

Cadwallader, K.R., Gnadt, T.A. and Jasso, L. 2007. Aroma components of chipotle peppers. In *Chemistry and Flavor of Hispanic Foods.* Tunick, M. and Gonzales de Mejia, E. (Eds), ACS Symposium Series 946, American Chemical Society, Washington, DC, pp. 57-66.

Lecona-Uribe, S., Loarca-Piña, G., Arcila-Lozano, C. and Cadwallader, K.R. 2007. Chemical characterization of *Lippa graveolens* Kunth and its comparison to *Origanum vulgare* and *Origanum laevigatum herrenhausen.* In *Chemistry and Flavor of Hispanic Foods.* Tunick, M. and Gonzales de Mejia, E. (Eds), ACS Symposium Series 946, American Chemical Society, Washington, DC, pp. 45-56.

Lozano, P.R., Cadwallader, K.R. and Gonzales de Mejia, E. 2007. Identification of characteristic aroma components of Mate (*Ilex paraguariensis*) tea. In *Chemistry and Flavor of Hispanic Foods.* Tunick, M. and Gonzales de Mejia, E. (Eds), ACS Symposium Series 946, American Chemical Society, Washington, DC, 143-152.

Cadwallader, K.R., Drake, M.A., Carunchia-Whetstine, M.E. and Singh, T.J. 2006. Characterisation of Cheddar cheese flavour by sensory directed instrumental analysis and model studies. In *Flavour Science: Recent Trends.* Bredie, W.P. and Peterson, M.A. (Eds.), Developments in Food Science 43, Elsevier, New York, pp. 157-160.

Jarunrattanasri, A., Cadwallader, K.R., Theerakulkait, C. 2005. Aroma and amino acid composition of hydrolyzed vegetable protein from rice bran. In *Process and Reaction Flavors*, Weerasinghe, D.K. (Ed.), ACS Symposium Series 905, American Chemical Society, Washington, DC, pp. 83-97.

Cadwallader, K.R., Benitez, D., Pojjanapimol, S., Syriyaphan, O. and Singh, T. 2005. Characteristic aroma components of the cilantro mimics. In. *Natural Flavors and Fragrances*, Frey, C. and Rouseff, R. (Eds.), ACS Symposium Series 908, American Chemical Society, Washington, DC, pp. 117-128.

Cadwallader, K.R. 2005. Flavor and volatile metabolism in produce. Chapter 6. In *Produce Degradation: Pathways and Prevention,* Lamikanra, O., Imam, S.H. and Ukuku, D.O. (Eds.), CRC Press LLC, Boca Raton, FL, pp 155-189.

Pojjanapimol, S., Chaiseri, S. and Cadwallader, K. 2004. Heat-induced changes in aroma components of holy basil (*Ocimum sanctum* L.). Ch. 15. In *Handbook of Flavor Characterization: Sensory analysis, chemistry, and physiology*, Diebler, K.D. and Delwiche, J. (Eds.), Marcel Dekker, Inc., New York, pp. 217-230.

Singh, T.K. and Cadwallader, K.R. 2004. Ways of measuring shelf-life and spoilage. Ch. 9. In *Understanding and Measuring the Shelf-life of Food*, Steele, R. (Ed.). CRC Press LLC, Boca Raton, FL, pp. 165-183.

Cadwallader, K.R., Lopez, J.R., Menke, S.D. and Surakarnkul, R. 2003. Aroma components of wines from Chardonel - A French-American hybrid grape. In *Nutraceutical Beverages. Chemistry, Nutrition, and Health Effects*, Shahidi, F. and Weerasinghe, D.K. (Eds.), ACS Symposium Series 871. American Chemical Society, Washington, DC, Chapter 27.

Karagül-Yüceer, Y., Drake, M.A. and Cadwallader, K.R. 2002. Aroma characterization of fresh and stored nonfat dry milk. In *Freshness and Shelf Life of Foods.* Cadwallader, K.R. and Weenen, H. (Eds.), ACS Symposium Series 836, American Chemical Society, Washington, DC, pp. 108-123.

Singh, T.K. and Cadwallader, K.R. 2002. The shelf life of foods: An overview. In *Freshness and Shelf Life of Foods.* Cadwallader, K.R. and Weenen, H. (Eds.), ACS Symposium Series 836. American Chemical Society, Washington, DC, pp. 2-21.

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Klein, B.P., Cadwallader, K.R., Chen, D., Khanna, P., Sullivan, C.L. and Weingartner, K.E. 2002. *Textured Vegetable Protein in the American Kitchen.* Illinois Center for Soy Foods, University of Illinois, University Board of Trustees, Urbana, IL.

Klein, B.P., Cadwallader, K.R., Chen, D., Khanna, P. and Weingartner, K.E. 2002. *Tofu in the American Kitchen.* Illinois Center for Soy Foods, University of Illinois, University Board of Trustees, Urbana, IL.

Miller, G.Y., Robert, M.J., Maghirang, R.G., Riskowski, G.L., Heber, A.J., and Cadwallader, K.R. Characterization of odor and gas emitting potential from mechanically ventilated deep and shallow pit swine finishing buildings in Illinois. In *Livestock Environment VI: Proceedings of the 6th International Symposium* (21-23 May 2001, Louisville, Kentucky, USA), Stowell, R.R., Bucklin, R. and Bottcher, R.W., Eds., May 21, 2001, pp. 664-670, ASAE Publication Number 701P0201.

Invited Talks

Cadwallader, K.R., Yin, Y., Hausch, B. and Chen, F. 2016. Assuring accuracy in the quantitation of the unstable odorant 2-acetyl-1-pyrroline in aromatic rice. ACS Symposium: *Flavor Stability: Chemical Changes in Flavor Molecules, Food-Flavor Matrix Interactions, Flavor Encapsulation.* 252nd Americal Chemical Society Meeting. Philadelphia, PA, August 21-25.

Cadwallader, K.R. 2016. Occurrence, formation and stabilization of the potent odorant 2-acetyl-1-pyrroline. Meeting of the Illinois Heartland Local Section of the American Chemical Society, Bloomington, IL, May 16, 2016.

Ickes, C. and Cadwallader, K.R. 2016. Characterizing aroma components of rum. ACS Symposium: *Flavor Chemistry of Alcoholic Beverages.* 251st American Chemical Society Meeting, San Diego, CA, March 13-17.

Cadwallader, K.R., Hausch, B. and Yin, Y. Quest for stability of 2-acetyl-1-pyrroline: The key odorant in aromatic rice and pandan. 17th Food Innovation Asia Conference 2015, Bangkok, Thailand, June 18-19.

Ickes, C. and Cadwallader, K.R. 2015. Understanding the effects of ethanol-flavor interactions on flavor perception in alcoholic beverages. ACS Symposium: *Challenges in Applied Flavor Sciences.* ACS National Meeting, Boston, MA, August 16-20, AGFD paper no. 297.

Pang, X., Wu, J. and Cadwallader, K.R. 2014. Identification of predominant contributors to off-odors formed in thermally processed melon juice. ACS Symposium: *Chemical Changes During Processing.* 248th American Chemical Society Meeting, San Francisco, CA, August 10-14. AGFD paper no. 177.

Cadwallader, K.R. and Lapsongphon, N. 2012. Identification of key odorants in fish sauce. US-Korea Conference (UKC) 2012, Los Angeles, August 8-11.

Fang, M. and Cadwallader, K.R. 2011. Convenient synthesis of stable deuterium labeled alkyl pyrazines for use in stable isotope dilution assays. ACS Symposium: *Recent Advances in Analysis of Food Flavors.* 242nd American Chemical Society Meeting, Denver, CO, August 28 – September 1, 2011, AGFD paper no. 11.

Hausch, B. and Cadwallader, K.R. 2010. Flavor chemistry of lemon-lime flavored carbonated beverages. ACS Symposium: *Symposium Honoring Prof. Terry E. Acree on the Occasion of His 70th Year.* 240th ACS National Meeting, Boston, MA, August 22-26, AGFD paper no. 40.

Lahne, J. and Cadwallader, K.R. 2010. Streamlined analysis of potent odorants in distilled alcoholic beverages. ACS Symposium: *Flavor Chemistry of Alcoholic Beverages.* 240th ACS National Meeting, Boston, MA, August 22-26, AGFD paper no. 106.

Cadwallader, K.R. and Kim, H. 2010. Flavor chemistry of fish sauce. ACS Symposium: *Fermented Foods.* 239th ACS National Meeting, San Francisco, CA, March 21-25, AGFD paper no. 112.

Cadwallader, K.R. 2010. Flavor chemistry of tree nuts. IFT Symposium: *Nuts: Phytochemicals, Health Effects and Flavor.* IFT Annual Meeting and Food Expo, Chicago, IL, July 17-20, paper no. 054-05.

Lozano, P.R. and Cadwallader, K.R. 2009. Effect of deamidation on binding of flavor compounds by dehydrated soy protein isolates. ACS Symposium: *Food Proteins and Food-based Protein Chemistry* 237th ACS National Meeting, Finley, J., organizer, Salt Lake City, UT, March 22-26.

Cadwallader, K.R. 2008. Sensory evaluation of aroma models for flavor characterization. *Symposium in Honor of Ronald Buttery G. Kenneth Recipient of the 2008 Spencer Award for Outstanding Achievement in Agricultural and Food Chemistry*, Kansas City Section, American Chemical Society, Kansas City, MO, Oct 27.

Cadwallader, K.R. 2008. AGFD and 100 years of chemical research on grains and legumes. ACS Symposium: *AGFD 100th Anniversary Symposium*, Brine, C. and Tunick, M.H., organizers, 236th ACS National Meeting, Philadelphia, PA, August 17-21.

Liu, Y and Cadwallader, K.R. 2008. Aroma components of "Carlos" muscadine (*Vitis rotundifolia* Michx) grape juice. ACS Symposium: Flavor and Health Benefits of Small Fruits, Rimando, A.M. and Qian, M.C., organizers, 236th ACS National Meeting, Philadelphia, PA, August 17-21.

Lozano, P.R. and Cadwallader, K.R. 2008. Flavor binding by soy proteins. ACS Symposium: Chemistry, Texture and Flavor of Soy, Cadwallader, K.R. and Chang, S. K. C. Chang, organizers, 236th ACS National Meeting, Philadelphia, PA, August 17-21.

Watcharananun, W., Cadwallader, K.R. and Huangrak, K. 2008. Predominant odorants in Thai desserts flavored by smoking with *tian op*, a traditional Thai scented candle. ACS Symposium: *Recent developments in flavor chemistry*, Cadwallader, K.R., organizer, 235th ACS National Meeting, New Orleans, LA, April 6-10.

Cadwallader, K.R. Lozano, P.R. and Kim, H. 2007. Challenges in the isolation and identification of key aroma components of complex functional foods. ACS Symposium: *Food Bioactives and Nutraceuticals: Production, Chemistry, Analysis and Health Effects: Symposium Honoring Fereidoon Shahidi*, Rimando, A.M., organizer. 234th ACS National Meeting, Boston, MA, August 19-23, AGFD Paper 92.

Cook, S., Cadwallader, K.R. and Kim, H. 2007. Identification of key thermally-derived odorants in cooked chicken liver. ACS Symposium: *Thermal Generation of Flavors and Off-flavors,* Ho, C.-T., Raghavan, S., McGorrin, R.J., organizers. 234th ACS National Meeting, August 19-23. AGFD Paper 206.

Cadwallader, K.R., Lorjaroenphon, Y., Kim, H. and Lee, S-Y. 2007. Evaluation of key odorants in chipotle pepper by quantitative analysis, calculation of odor-activity values and omission studies. 8th Wartburg Symposium on Flavor Chemistry & Biology, Eisenach, Germany, February 28 – March 2.

Cadwallader, K.R. 2007. Protein-based functional beverages and flavoring considerations. In IFT Symposium: *Functional Beverages and Human Health.* Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 28 - August 1, Session-Paper 174-02.

Cadwallader, K.R. 2006. Chemical and sensory characterization of food aroma: An academic approach. Meeting of the Society of Flavor Chemists, Chicago, IL, October 19.

Cadwallader, K.R. 2006. Flavor chemistry of fish and shellfish. ACS Symposium: Molecular Cuisine. (Tunick, M.H., Risch, S.J. and Johnson, S.C., organizers). 232nd ACS National Meeting, San Francisco, CA, September 10-14, AGFD paper 61.

Cadwallader, K.R. and Zhou, Q. 2006. Characteristic aroma components of tree nuts. ACS Symposium: Nutrition and Sensory Quality of Nuts. (Pattee, H.E.; Shahidi, F.; Yu, L., organizers). 331st ACS National Meeting, Atlanta, GA, March 26 – March 30, AGFD paper 40.

Cadwallader, K.R. 2006. Modern techniques to characterize complex food flavor systems. International Symposium on *Functional Foods*, ATAM (Nutrition Society of Mexico) and Institute of Food Technologists, Mexico City, Mexico, August 17.

Rotsachakul, P., Cadwallader, K.R. and Chaiseri, S. 2005. Comparison of DHS and solvent extraction-SAFE for GCO-dilution analysis of key aroma components of Thai fried chili paste. ACS Symposium: *Aroma-active Compounds in Foods and Reaction Flavors: Formation, Measurement and Stability.* (Ho, C.-T., Weerasinghe, D.K. and Cadwallader, K.R., organizers). 330th ACS National Meeting, Washington, DC, August 28 – September 1, AGFD paper 9.

Cadwallader, K.R., Bordignon, J.R., Wang, P. and Sugiharto, R. 2005. Identification of odorants responsible for off-flavor of corn gluten meal. ACS Symposium: *Grain-based Products: Health and Flavor, and Safety Aspects* (Cadwallader, K.R., Schieberle, P. and Morello, M.J., organizers). 330th ACS National Meeting, Washington, DC, August 28 – September 1, AGFD paper 167.

Cadwallader, K.R., Drake, M.A., Carunchia-Whetstine, M.E. and Singh, T.J. 2005. Characterisation of Cheddar cheese flavour by sensory directed instrumental analysis and model studies. 11th Weurman Flavour Research Symposium, Roskilde, Denmark, June 21-24.

Gnadt, T.A., Jasso, L. and Cadwallader, K.R. 2005. Characteristic aroma components of chipotle peppers. ACS Symposium: *Chemistry and Flavor of Hispanic Foods* (de Mejia, E. and Tunick, M.H., organizers). 229th ACS National Meeting, San Diego, CA, March 13-17.

Lozano, P.R., Nicolalde, C.L. and Cadwallader, K.R. 2005. Identification of characteristic aroma components of mate (*Ilex paraguariensis*) tea. ACS Symposium: *Chemistry and Flavor of Hispanic Foods* (de Mejia, E. and Tunick, M.H., organizers). 229th ACS National Meeting, San Diego, CA, March 13-17.

Lecona-Uribe, S., Loarca-Piña, G., Arcila-Lozano, C.C. and Cadwallader, K.R. 2005. Chemical composition of essential oil from Mexican orégano. ACS Symposium: *Chemistry and Flavor of Hispanic Foods* (de Mejia, E. and Tunick, M.H., organizers). 229th ACS National Meeting, San Diego, CA, March 13-17.

Cadwallader, K.R. Importance of sample preparation technique on the composition of aroma extracts from fresh herbs. ACS Sympsosium: *Challenges in Herb Research – Chemical Analysis and Quality* (Ho, C.T. and Hwang, L.S., organizers). 227th ACS National Meeting, Anaheim, CA, March 28-April 1, 2004.

Cadwallader, K.R., Jarunrattanasri, A. and Theerakulkait, C. 2003. Aroma components of HVP made from rice bran protein. ACS Symposium: *Processed/Reaction Flavors, Ingredients and Intermediate Reactions.* (Weerasinghe, D.K. and Sucan, M.K., organizers). 226th ACS National Meeting, New York, NY, September 7-11.

Cadwallader, K.R. and Pojjanapimol, S. 2003. Characteristic aroma components of the cilantro mimics. ACS Symposium: *Natural Flavor and Fragrance Ingredients: Chemistry, Production and Application.* (Frey, C. and Rouseff, R.L., organizers). 226th ACS National Meeting, New York, NY, September 7-11.

Cadwallader, K.R. Relating Cheddar cheese flavor to chemical components. IFT Symposium: *Flavor Chemistry of Dairy Products.* (Drake, M.A., organizer). Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 12-16, #52-5.

Cadwallader, K.R., Pojjanapimol, S. and Chaiseri, S. 2002. Heat-induced changes in holy basil aroma components. ACS Symposium: *Challenges in the Characterization of Flavor Compound.* (Deibler, K.D. and Delwiche, J., organizers). 224th ACS National Meeting, Boston, MA, August 18-22.

Cadwallader, K.R., Zhou, Q. and Huda, M.S. 2002. Measuring flavor release from microencapsulants under controlled environmental conditions. ACS Symposium: *Controlled Release Systems for Foods.* (Shefer, S. and Yildiz, M.E., organizers). 223rd ACS National Meeting, Orlando, FL, April 7-11.

Cadwallader, K.R., Brehart, D.M., Schmidt, S.J. and Huda, M.S. 2002. Volatile release from a sucrose encapsulated flavoring. IFT Symposium: *Use of Carbohydrates for Encapsulation and Sustained Release.* (Obanni, M. and Asquith, T.N., organizers). Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #67-1.

Cadwallader, K.R., Lopez, J.R. and Menke, S.D. 2001. Aroma components of wines from Chardonel, a French-American hybrid grape variety. ACS Symposium: *Chemistry and Flavor of Nutritional Beverages.* (Shahidi, F. and Weerasinghe, D., organizers). 222nd ACS National Meeting, Chicago, IL, August 26-31.

Guthrie, B. and Cadwallader, K.R. 2001. Measuring flavor retention in dry food systems. IFT Symposium: *Developments in the Analysis of Complex Flavor Systems*. (Cadwallader, K.R. and McGorrin, R.J., organizers). Annual Meeting of the Institute of Food Technologies, New Orleans, LA, June 23-27, #76-2.

Cadwallader, K.R. 2001. Use of SPME Technology to Evaluate Antioxidant Effectiveness. Symposium: *Antioxidant Analysis*, (Engeseth, N.J., organizer). 92nd AOCS Annual Meeting and Expo, Minneapolis, MN, May 13-16.

Karagul-Yuceer,Y., Drake, M.A. and Cadwallader, K.R. 2001. Aroma characterization of fresh and stored nonfat dry milk. ACS Symposium: *Freshness and Shelf-life of Foods.* (Weenen, H. and Cadwallader, K.R., organizers). 222nd ACS National Meeting, Chicago, IL, August 26-30.

Cadwallader, K.R. 2000. Flavor Chemistry of Saffron. Symposium: *Carotenoid-Derived Aroma Compounds.* (Winterhalter, P. and Rouseff, R., organizers). 219th ACS National Meeting, San Francisco, CA, March 26-30, paper #46 AGFD.

Cadwallader, K.R. 1999. Sample dilution analysis for identification of aroma-impact components of edible oils. ACS Symposium: *Advances in Gas Chromatography/Olfactometry.* (Leland, J., Acree, T.E., organizers). 218th ACS National Meeting, New Orleans, LA, August 22-26.

Cadwallader, K.R. and Shahidi, F. 1999. Identification of potent odorants in seal blubber oil by direct thermal desorption-gas chromatography-olfactometry. ACS Symposium: *Chemistry and Nutrition of Highly Unsaturated Fatty Acids.* (Shahidi, F. and Finley, J.W., organizers). 217th ACS National Meeting, Anaheim, CA, March 21-25.

Cadwallader, K.R. and Howard, C.L. 1997. Analysis of aroma-active compounds in light-activated milk. ACS Symposium: *Challenges in the Isolation and Characterization of Flavor Compounds.* (Mussinan, C.J. and Morello, F., organizers). 214th National Meeting of the American Chemical Society, Las Vegas, NV, September 7-11.

Baek, H.-H. and Cadwallader, K.R. 1996. Character-impact aroma compounds of crustacea. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods.* (Shahidi, F. and Cadwallader, K.R., organizers). 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Cha, Y.J., Lee, G.H. and Cadwallader, K.R. 1996. Aroma-active compounds in salt-fermented anchovy. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods.* (Shahidi, F. and Cadwallader, K.R., organizers). 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Wang, H.-J., Cadwallader, K.R., Baek. H.H., D'Abramo, L.R. and Sullivan, J.A. 1996. Manipulating the flavor of crustacea using postharvest seawater acclimation. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods.* (Shahidi, F. and Cadwallader, K.R., organizers). 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Cadwallader, K.R. and Howard, C.L. 1996. Characterization of saffron flavor by aroma extract dilution analysis. ACS Symposium: *Spices: Flavor Chemistry and Antioxidant Properties.* (Risch, S.J. and Ho, C.T., organizers). 211th National Meeting of the American Chemical Society, New Orleans, LA, March 24-28.

Cadwallader, K.R., Baek, H.H., Chung, H.Y. and Moody, M.W. 1993. Contribution of lipid-derived components to the flavor of alligator meat. ACS Symposium: *Lipids and Food Flavors.* (Ho, C.T. and Hoffman, T.G., organizers). 205th National Meeting of the American Chemical Society, Denver, CO.

Braddock, R.J. and Cadwallader, K.R. 1993. Bioconversion of citrus d-limonene. Symposium paper presented at the 206th National Meeting of the American Chemical Society, Chicago, IL.

Braddock, R.J. and Cadwallader, K.R. 1991. Citrus by-products manufacture for food use. Symposium paper presented at the 52nd Annual Meeting of the Institute of Food Technologists, Dallas, TX, June 1-5.

Volunteered Abstracts

Kelley, L. and Cadwallader, K.R. 2016. Identification and quantitation of potent odorants in spearmint oils. 11th Wartburg Syposium on Flavor Chemistry & Biology, Eisenach, Germany, June 21-24.

McKenna, S. and Cadwallader, K.R. 2016. Identification and characterization of mint lactones: Trace-level odor-important compounds in peppermint oil. IFT Annual Meeting and Food Expo, Chicago, IL, July 17-19, paper no. PO2-020

Ickes, C., Lee, S. and Cadwallader, K.R. Novel creation of rum flavor lexicon through the use of web-based material. IFT Annual Meeting and Food Expo, Chicago, IL, July 17-19, paper no. P03-111

Yu, L., Thomas, L., Yin, L, Jerrell, J., Cadwallader, K. and Schmidt, S. 2015. Impact on sucrose crystal composition and chemistry on its thermal behavior. IFT Annual Meeting and Food Expo, Chicago, IL, July 11-14, paper no. 046-093.

Yu, L., Thomas, L., Yin, L, Jerrell, J., Cadwallader, K. and Schmidt, S. 2015. Impact on sucrose crystal composition and chemistry on its thermal behavior. IFT Annual Meeting and Food Expo, Chicago, IL, July 11-14, paper 046-093.

Cadwallader, K.R. and Fang, M.-C. 2014. A novel and attractive method for the stabilization of the potent odorant 2-acetyl-1-pyrroline. 14th Weurman Flavor Research Symposium, Cambridge, UK, September 15-19.

Donovan, J.D., Cadwallader, K.R. and Lee, Y. 2014. Improvement of protein microcapsule structure and core retention through the use of inulin. IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 181-01.

Erten, E. and Cadwallader, K.R. 2014. Characterization of aroma components of raw, dry roasted, and oil roasted almonds using solvent assisted flavor evaporation (SAFE) and aroma extract dilution analysis (AEDA). IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 206-26.

Lu, Y., Lee, J., Thomas, L., Jerrell, J., Cadwallader, K.R. and Schmidt, S.J. 2014. Investigating the thermal decomposition differences between beet and cane sugars using HPLC. IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 050-07.

Rosales, E., Cadwallader, K.R. and Engeseth, N. 2014. Effect of ultrasonic application on fatty acid composition and aroma compounds of dark chocolate. IFT Annual Meeting and Food Expo, New Orleans, LA, June 20-25, paper 206-94.

Burnsides, E. and Cadwallader, K. 2012 Comparison of aroma compounds in rums and rum ethers. IFT Annual Meeting and Food Expo, Las Vegas, NV, paper no. 193-28.

Su, C., Padua, G. and Cadwallader, K. 2012. Zein applications in flavor encapsulation. IFT Annual Meeting and Food Expo, Las Vegas, NV, paper no. 228-23.

Cadwallader, K.R., Hausch, B. and Lorjaroenphon, Y. 2011. Identification, characterization and relative aroma impact of 2,3-dehydro-1,8-cineol in lemon-lime flavored carbonated beverages. 13th Weurman Flavor Research Symposium, Zaragoza, SPAIN, September 26-30.

Suppavorasatit, I., deMejia, E. and Cadwallader, K.R. 2011. Effect of enzymatic deamidation of soy protein isolate by protein-glutaminase on the functional properties of the protein. IFT Annual Meeting and Food Expo, New Orleans, LA, June 11-14, paper no. 236-57.

Lapsongphon, N., Yongsawatdigul, J. and Cadwallader, K.R. 2011. Aroma characterization of premium fish sauce. IFT Annual Meeting and Food Expo, New Orleans, LA, June 11-14, paper no. 236-61.

Tinchan, P., Chaiseri, S. and Cadwallader, K.R. 2011. Quantification of δ-lactones in canned coconut milk by stable isotope dilution assay-GC-MS. IFT Annual Meeting and Food Expo, New Orleans, LA, June 11-14, paper no. 236-64.

Lorjaroenphon, Y. and Cadwallader, K.R. 2011. Identification of potent odorants in cola flavored carbonated beverages. IFT Annual Meeting and Food Expo, New Orleans, LA, June 11-14, paper no. 236-73.

Gao, Y., Cadwallader, K.R. and Schmidt, S.J. 2011. Investigating the chemical and physical stability of glass encapsulated spray-dried flavors stored below their glass transition temperatures. IFT Annual Meeting and Food Expo, New Orleans, LA, June 11-14, paper no. 290-18

Lee, H., Kim, H., Cadwallader, K.R. and Feng, H. 2011. Analysis of aroma compounds of apple cider treated by thermal-pasteurization, manosonication, thermosonication, and manothermosolication. IFT Annual Meeting and Food Expo, New Orleans, LA, June 11-14, paper no. 293-14.

Cadwallader, K.R. 2010. Potent odorants in soymilk as affected by processing and storage. *Nutraceuticals, Ingredients, Challenges, Formulations, Flavours and Health Effects.* International Conference, Exhibition and Short Course on Nutraceuticals and Functional Foods, Denpasar, Bali, Indonesia, October 11 – 12, paper no. 92.

Genthner, E.R. and Cadwallader, K.R. 2010. Characterizing aroma components of fresh-cut watermelon. 9th Wartburg Symp. on Flavor Chemistry & Biology, Eisenach, Germany, April 13-16.

He, Y., Garces, M., Toshkov, S., Kim, H., Cadwallader, K.R., Engeseth, N., Parker, T. and Jackson, M. 2010. Impact of irradiation processing on papaya nectar flavor. IFT Annual Meeting and Food Expo, Chicago, IL, July 17-20, paper no. 188-11

Kim, H. and Cadwallader, K.R. 2010. Comparison of key odorants between commercial aseptic Korean and American soy milks. IFT Annual Meeting and Food Expo, Chicago, IL, July 17-20, 188-05

Tinchan, P., Chaiseri, S. and Cadwallader, K.R. 2010. Changes in the volatile profile of canned coconut milk during storage. IFT Annual Meeting and Food Expo, Chicago, IL, July 17-20, 188-14.

Cadwallader, K.R. 2009. Flavor challenges in functional and nutraceutical beverages. International Conference and Exhibition on Nutraceuticals and Functional Foods 2009, International Society for Nutraceuticals & Functional Foods, San Francisco, CA, October 31-November 4, paper 87.

LaFond, S., Cadwallader, K. and Artz, W. 2009. The formation of 4-HNE in frying oils: A controlled study using a French fry model. 110th AOCS Annual Meeting & Expo, Orlando, FL, May 3-6.

Choi, I.-S., Kim, H. and Cadwallader, K.R. 2009. Comparison of the aroma components between dairy and soy yogurt. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 6-9, #024-02.

Lorjaroenphon, Y., Moy, M., Rosales, E. and Cadwallader, K.R. 2009. Identification of the key aroma components of fresh and baked Bartlett pear. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 6-9, #057-15.

Lozano, P.R. and Cadwallader, K.R. 2009. Effect of nonenzymatic deamidation of soy protein isolate on the binding of selected flavor compounds. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 6-9, #024-12

Lozano, P. and Cadwallader, K. 2008. Binding interactions of selected sulfur compounds with soy protein isolate under controlled relative humidity. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 28 – July 1, #006-12.

Carrasco Hernandez, G. and Cadwallader, K.R. 2008. Headspace reduction by different terpenoids of the main volatile sulfur compounds responsible for garlic breath. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 28 – July 1, #006-24.

Waimaleongora-Ek, P, Herrera Corredor, J., Prinyawiwatkul., W., Suvanich, V. and Cadwallader, K. Sensory attributes driving acceptance and purchase intent of watermelon juice containing no added sugar. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 28 – July 1, #138-12

Cadwallader, K.R. and Puangpraphant, S. 2008. Changes in the aroma components of pecans during roasting. 12th Weurman Flavour Research Symposium, Interlaken, Switzerland, July 1-4

Kim, H., Cadwallader, K.R. and Kido, H. 2008. Effect of commercial rosemary extract powders on formation of lipid-derived odorants in cooked pork sausages during cold storage. 235th ACS National Meeting, New Orleans, LA, April 6-10.

Carrasco, G. and Cadwallader, K.R. 2007. Mechanism of action of menthone in ameliorating oral cavity malodor caused by consumption of garlic. 234th ACS National Meeting, Boston, MA, August 19-23.

Lozano, P.R., Miracle, E.R., Krause, A.J. and Drake, M.A. 2007. Effect of cold storage and packaging material on the major aroma components of sweet cream butter. 234th ACS National Meeting, Boston, MA, August 19-23.

Gnadt, T.A., Lee, S.-Y. and Cadwallader, K.R. 2007. Effects of commercial masking agents and lactones on soymilk aroma and flavor. Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 28 - August 2, #4-224.

Hong, L., Sajuti, S.C. and Cadwallader, K.R. 2007. Identification of characteristic aroma components of Pomegranate (Punica granatum). Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 28 - August 2, #3-187

Mosca, C., Esgro, S. and Cadwallader, K.R. 2007. Flavor Characterization of unrefined shea butters. American Oil Chemists' Society's (AOCS) 98th Annual Meeting and Expo, Quebec City, QC, Canada, May 13-16.

Andrae, L.M., Cadwallader, K.R. and Engeseth, N.J. 2006. Flavor changes of stored dark chocolate by dynamic headspace sorption – gas chromatography/mass spectrometry. Annual Meeting of the Institute of Food Technologists, Orlando, FL, June 24-28, #39F-12.

Lozano, P. and Cadwallader, K.R and Drake, M.A. 2006. Instrumental and sensory characterization of heat-induced odorants in aseptic packaged soymilk. Annual Meeting of the Institute of Food Technologists, Orlando, FL, June 24-28, #39F-12.

Gnadt, T.A., Jasso, L. and Cadwallader, K.R. 2005. Comparison of direct solvent extraction and dynamic headspace sampling for the analysis of characteristic aroma components of chipotle pepper. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, July 16-20, #17D-34.

Klein, B.P., Khanna, P., Cadwallader, K.R. and Endres, J.M. 2005. Reducing fat in school lunches by using soy-enhanced entrees. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, July 16-20, #70E-27.

Nowak, M.C., Singh, T.K. and Cadwallader, K.R. 2005. Identification of potent odorants in commercial hickory and mesquite liquid smoke. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, July 16-20, #17D-36.

Song, H. and Cadwallader, K.R. 2005. Aroma components of American country-style ham. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, July 16-20, #17D-35

Yu, X., Song, Y.S. and Cadwallader, K.R. 2005. Identification of the characteristic aroma components of commercial dried California bay leaves (*Laurus nobilis* L.). Annual Meeting of the Institute of Food Technologists, New Orleans, LA, July 16-20, #17D-19.

Zhou, Q. and Cadwallader, K.R. 2005. Application of IGC for measurement of binding of volatile flavor compounds by soy proteins in low-moisture food systems. 229th ACS National Meeting, San Diego, CA, March 13-17.

Bohn, D.M. Cadwallader, K.R. and Schmidt, S.J. 2004. Kinetic analysis of benzaldehyde release in artificial cherry Durarome® upon humidification using dynamic vapor sorption-fast gas chromatography-flame ionization detection. Annual Meeting of the Institute of Food Technologists, Las Vegas, NV, July 12-16, #114B-27.

Lopez, J.R., Cadwallader K.R. and Menke, S.D. 2004. Menke Aroma-impact components of Illinois Frontenac wine. Annual Meeting of the Institute of Food Technologists, Las Vegas, NV, July 12-16, #83D-3.

Singh, T.K., Young, N.D., Drake, M.A., Cadwallader, K.R. 2004. Production of a bitter peptide from casein and its sensory characterization. Annual Meeting of the Institute of Food Technologists, Las Vegas, NV, July 12-16, #17B-17.

Ugarte-Romero, E.E. Feng, H, Martin, S.E. and Cadwallader, K.R. 2004. Power ultrasound as an alternative for apple cider pasteurization. Annual Meeting of the Institute of Food Technologists, Las Vegas, NV, July 12-16, #32-10.

Yoon, Y., Carunchia-Whetstine, M.E., Friedeck, K.G., Drake, M.A. and Cadwallader, K.R. 2004. Characterization of aroma-active compounds in fresh sweet cherry cultivars. Annual Meeting of the Institute of Food Technologists, Las Vegas, NV, July 12-16, #114B-3.

Zhou, Q. and Cadwallader, K.R. 2004. Investigation of flavor-soy protein interactions under controlled relative humidity by inverse gas chromatography (IGC). Annual Meeting of the Institute of Food Technologists, Las Vegas, NV, July 12-16, # 114B-2.

Brisske, L.K., Lee, S.Y., Klein, B.P. and Cadwallader, K.R. 2003 Consumer ratings of energy bars: Should the bar be raised? Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 12-16, #12-2.

Bohn, D.M., Cadwallader, K.R. and Schmidt, S.J. 2003. Measurement of volatile release from an amorphous carbohydrate matrix as a function of relative humidity using dynamic vapor sorption technology. Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 12-16, #12-2.

Singh, T.K., Drake, M.A. and Cadwallader, K.R. 2003. Characterization of aroma components of Muenster cheese by aroma extract dilution and headspace dilution analyses. Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 12-16, #71-3.

Suriyaphan, O., Zhou, Q. and Cadwallader, K.R. 2003. Identification of key aroma components of *Piper betel* L. leaf. Annual Meeting of the Institute of Food Technologists, Chicago, IL, July 12-16, #45C-16.

Drake, M.A., Karagul-Yuceer, Y., Civille, G.V., Cadwallader, K.R. and Tong, P.S. 2002. Development of a descriptive sensory language for milk powders. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #100D-2.

Karagul-Yuceer, Y., Drake, M.A. and Cadwallader, K.R. 2002. Evaluation of the character-impact odorants in nonfat dried milk by sensory studies on model mixtures. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #12-10.

Karagul-Yuceer, Y., Vlahovich, K., Drake, M.A. and Cadwallader, K.R. 2002. Compounds responsible for musky off-odor of rennet casein. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #15B-12.

Karagul-Yuceer, Y, Cadwallader, K.R. and Drake, M.A. 2002. Odor-active compounds of liquid whey. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #61B-4.

Pojjanapimol, S., Chaiseri, S. and Cadwallader, K.R. 2002. Aroma components of distilled essential oil of Holy basil. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #61B-12.

Zhou, Q., Cadwallader, K.R. and Guthrie, B.D. 2002. Simultaneous measurement of permeability of aroma compound mixtures through polymer packaging films. Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 15-19, #100B-8.

Benitez, D. and Cadwallader K.R. 2001. Aroma-active components of Vietnamese coriander (*Polygonum odoratum*). Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #88A-14.

Benitez, D., Kappes, S. and Cadwallader, K.R. 2001. Aroma of dried peaches. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #44C-34.

Chen, D., Andrae, L., Suriyaphan, O., Weingartner, K. and Cadwallader, K. 2001. Characteristic aroma components of vegetable soybean (edamame). Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #88A-17.

Drake, M.A., Gerard, P.D., Wright, S., Cadwallader, K.R. and Civille, G.V. 2001. Cross validation of a sensory language for Cheddar cheese. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #73G-19.

Grady, J., Klein, B.P. and Cadwallader, K.R. 2001. Bitterness and astringency thresholds for soy isoflavones. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #98-2.

Karagul-Yuceer, Y, Cadwallader, K.R. and Drake, M.A. 2001. Identification of volatile off-flavor compounds of nonfat dried milks. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #88A-3.

Suvanich, V., Maciel-Pedrote, I., Cadwallader, K.R., Moser, E.B., Thongthai, C. and Prinyawiwatkul, W. 2001. Aroma indices of Southeast Asian fish sauces. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #73F-16.

Suvanich, V., Maciel-Pedrote, I., Cadwallader, K.R., Moser, E.B., Thongthai, C. and Prinyawiwatkul, W. 2001. Developnebt if fish sauces quantitative descriptive sensory aroma profile. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #73G-34.

Zhou, X., Winterstein, C. and Cadwallader, K.R. 2001. Aroma of buckwheat honey. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #88A-12.

Zhu, L., Robbins, K. and Cadwallader, K.R. 2001. Effects of soy protein isolate on headspace aroma components of cooked ground beef patties. Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 23-27, #88A-18.

Drake, M.A., McIngvale, S.C., Cadwallader, K.R. and Civille, G.V. 2000. Development of a descriptive language for Cheddar cheese. Book of Abstracts, Annual Meeting of the Institute of Food Technologists, Dallas, TX, June 10-14, #11-3.

Suriyaphan, O., Cadwallader, K.R. and Drake, M.A. 2000. Lipid oxidation of deoiled soy lecithin in fermented milk systems. Book of Abstracts, Annual Meeting of the Institute of Food Technologists, Dallas, TX, June 10-14, #14B-32.

Cadwallader, K.R. and Thongthai, C. 2000. Comparison of aroma-active headspace components of three types of fish sauces. Book of Abstracts, Annual Meeting of the Institute of Food Technologists, Dallas, TX, June 10-14, #36-3.

Chen, X.Q., Drake, M.A. and Cadwallader, K.R. 2000. Characteristic aroma components of English Farmhouse Cheddar Cheeses. Book of Abstracts, Annual Meeting of the Institute of Food Technologists, Dallas, TX, June 10-14, #78C-4.

Karagul, Y., Drake, M.A. and Cadwallader, K.R. 2000. Aroma-active components of nonfat dry milk powders. Book of Abstracts, Annual Meeting of the Institute of Food Technologists, Dallas, TX, June 10-14, #78C-6.

Cadwallader, K.R. and Jung, J. 1999. Evaluation of warmed-over flavor in chicken wings by sensory and instrumental methods. 60th Annual Meeting of the Institute of Food Technologists, Chicago, IL, #50A-13, p. 122.

Wu, Y.F., Baek, H.H. and Cadwallader, K.R. 1999. Development of a meat-like process flavoring from soybean-based hydrolyzed vegetable protein. 60th Annual Meeting of the Institute of Food Technologists, Chicago, IL, #50A-9, p.121.

Suriyaphan, O., Cadwallader, K.R. and Drake, M.A. 1999. Lecithin associated off-flavors in fermented dairy products—A model study. 60th Annual Meeting of the Institute of Food Technologists, Chicago, IL, #17-2, p. 31.

Cadwallader, K.R. and Suriyaphan, O. 1998. Aroma-impact components of loroco. 59th Annual Meeting of the Institute of Food Technologists, Atlanta, GA, #45-2, p. 97.

Surakarnkul, R. and Cadwallader, K.R. 1998. Release of glycosidically-bound aroma compounds during fermentation of muscadine juice. 59th Annual Meeting of the Institute of Food Technologists, Atlanta, GA, #72-18, p. 180.

Suriyaphan, O., Drake, M.A., and Cadwallader, K.R. 1998. Identification of volatile off-flavors in reduced fat cheeses containing lecithin. 59th Annual Meeting of the Institute of Food Technologists, Atlanta, GA, #72-31, p. 183.

Baek, H.H. and Cadwallader, K.R. 1997. Free and glycosidically bound volatile aroma compounds in muscadine grape juice. 58th Annual Meeting of the Institute of Food Technologists, Orlando, FL, #23D-6, p.63.

Cai, M. and Cadwallader, K.R. 1997. (Z)-3-Nonenal and (Z,Z)-3,6-nonadienal: Character-impact aroma compounds in watermelon. 58th Annual Meeting of the Institute of Food Technologists, Orlando, FL, #23D-9, p.64.

Cha, Y.J., Kim, H., and Cadwallader, K.R. 1997. Aroma-active compounds in Kimchi during aging. 58th Annual Meeting of the Institute of Food Technologists, Orlando, FL, #23D-13, p.65.

Baek, H.H., Kim, C.J., Ahn, H.S., Nam, H.S., and Cadwallader, K.R. 1997. Aroma extract dilution analysis of beef-like process flavor from extruded enzymatically hydrolyzed soybean protein. 58th Annual Meeting of the Institute of Food Technologists, Orlando, FL, #23D-24, p.67.

Fu, S.G., Baek, H.H., and Cadwallader, K.R. 1997. Comparison of aroma-active compounds extracted from blue crab meat with processing by-products hydrolysate using aroma extract dilution analysis. 58th Annual Meeting of the Institute of Food Technologists, Orlando, FL, #23D-25, p.67.

Aryana, K.J., Cadwallader, K.R., and Haque, Z.U. 1996. Isolation of flavor volatiles in lowfat Cheddar cheese by vacuum simultaneous distillation-solvent extraction. 73rd Annual Meeting of the Southern Branch, American Dairy Science Association, February 3-7.

Baek, H.H. and Cadwallader, K.R. 1996. Evaluation of the enzymatic hydrolysis of defatted soybean meal by response surface methodology. 57th Annual Meeting of the Institute of Food Technologists, New Orleans, LA, #53A-16, p.120.

Baek, H.H., Cadwallader, K.R., Marroquin, E., and Silva, J.L. 1996. Gas chromatography/olfactometric analysis of muscadine grape aroma. 57th Annual Meeting of the Institute of Food Technologists, New Orleans, LA, #34-7, p.74.

Cadwallader, K.R. 1996. Potent odorants in hickory and mesquite smokes and liquid smoke extracts. 57th Annual Meeting of the Institute of Food Technologists, New Orleans, LA, #34-6, p. 74.

Cadwallader, K.R., Baek, H.H., Chang-Lee, G.-H. 1996. Aroma extract dilution analysis of cooked freshwater prawn. 57th Annual Meeting of the Institute of Food Technologists, New Orleans, LA, #80D-20, p.197.

Cha, Y.J., Kim, E.J., Baek, H.H., and Cadwallader, K.R. 1996. Development of a flavoring agent from sea mussel. 57th Annual Meeting of the Institute of Food Technologists, New Orleans, LA, #80D-22, p.197.

Kim, H.R., Ahn, C.B., Jung, K.J., Baek, H.H., Cadwallader, K.R. 1996. Characterization and utilization of waste water from oyster processing plants as flavoring agents. 57th Annual Meeting of the Institute of Food Technologists, New Orleans, LA, #80D-21, p.197.

Aryana, K.J., Cadwallader, K.R., and Haque, Z.U. 1996. Development of methodology for measuring+ flavor volatiles in Cheddar cheese made with and without fat replacers. International Conference on Food Science and Technology of The Fine Particle Society, Chicago, IL, August 24-26.

Cha, Y.J., Baek, H.H., and Cadwallader, K.R. 1995. Volatile flavor components in oyster hydrolysate produced with protease. 56th Annual Meeting of the Institute of Food Technologists, Anaheim, CA, #54E-16, p.166.

Conner, T.M., McGregor, J.U., and Cadwallader, K.R. 1995. Identifying flavor components in light-activated milk. J. Dairy Sci. 48(supplement 1): 317.

Baek, H.H. and Cadwallader, K.R. 1994. Comparison of volatile compounds in flavor concentrates produced from crawfish processing by-products with and without protease treatment. 55th Annual Meeting of the Institute of Food Technologists, June 25-29, Atlanta, GA, #79C-5, p.244.

Cadwallader, K.R., Tan, Q., Chen, F., and Meyers, S.P. 1994. Identification of character-impact aroma compounds in spiny lobster (*Panulirus argus*) tail meat. 55th Annual Meeting of the Institute of Food Technologists, June 25-29, Atlanta, GA, #7-7, p.13.

Chung, H.Y. and Cadwallader, K.R. 1994. Comparison of the aroma of three different extracts prepared from blue crab (*Callinetes sapidus*) processing by-product. 55th Annual Meeting of the Institute of Food Technologists, June 25-29, Atlanta, GA, #79C-7, p.244.

Chung, H.Y., Chen, F., and Cadwallader, K.R. 1994. Evaluation of aroma of blue crab (*Callinetes sapidus*) claw and lump meats. 55th Annual Meeting of the Institute of Food Technologists, June 25-29, Atlanta, GA, #79C-6, p.244.

Tan, Q. and Cadwallader, K.R. 1994. Biotransformation of ß-pinene by *Aspergillus cellulosae*. 55th Annual Meeting of the Institute of Food Technologists, June 25-29, Atlanta, GA, #24C-7, p.67.

Andrews, L.S. and Cadwallader, K.R. 1993. Microbial and sensory quality of ground ginger as affected by gamma irradiation. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #817, p.206.

Baek, H.H. and Cadwallader, K.R. 1993. Identification of characteristic volatile flavor components of roasted chicory by aroma extract dilution analysis. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #506, p.129.

Cha, Y.J. and Cadwallader, K.R. 1993. Volatile flavor components in Korean fermented fish paste. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #510, p.130.

Cha, Y.J., Baek, H.H., and Cadwallader, K.R. 1993. Comparison of DHS and SDE for the analysis of volatile compounds in fermented shrimp paste. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #511, p.131.

Chung, H.Y. and Cadwallader, K.R. 1993. Aroma extract dilution analysis of freshly picked cooked blue crab meat. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #563, p.144.

Chung, H.Y., Chou, C.-C., and Cadwallader, K.R. 1993. Changes in the volatile flavor constituents of coconut during roasting. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #504, p.129.

Yoo, S.K. and Cadwallader, K.R. 1993. Bioconversion of α- and ß-pinene. 54th Annual Meeting of the Institute of Food Technologists, July 10-14, Chicago, IL, Technical Program: Book of Abstracts, #516, p.132.

Baek, H.H. and Cadwallader, K.R. 1992. Enzymatic hydrolysis of crawfish waste for bioflavor production. Ninth International Biotechnology Symposium and Exposition, August 16-21, Crystal City, VA, Book of Abstracts, #472.

Baek, H.H., Pike, M.M., Bidner, T.D., Moody, M.W., and Cadwallader, K.R. 1992. Evaluation of the off-odor associated with wild alligator. 53rd Annual Meeting of the Institute of Food Technologists, June 20-24, New Orleans, LA. Book of Programs and Abstracts, #359, p.93.

Cha, Y.J., Baek, H.H., and Cadwallader, K.R. 1992. Volatile flavor components in concentrated snow crab extracts. 53rd Annual Meeting of the Institute of Food Technologists, June 20-24, New Orleans, LA, Book of Abstracts, #357, p.92.

Chung, H.Y. and Cadwallader, K.R. 1992. Comparison of volatile compounds in freshly picked crabmeat and crab processing waste. 53rd Annual Meeting of the Institute of Food Technologists, June 20-24, New Orleans, LA, Book of Abstracts, #358, p.93.

Kim, H.R., Baek, H.H., Cadwallader, K.R., Meyers, S.P., and Godber, J.S. 1992. Improved flavor extractibility from crab waste using the hepatopancreatic enzyme of crawfish, *Procambarus clarkii*. 53rd Annual Meeting of the Institute of Food Technologists, June 20-24, New Orleans, LA, Book of Abstracts, #360, p.93.

Cadwallader, K.R. and Braddock, R.J. 1990. Enzymatic hydration of limonene to α-terpineol. 51st Annual Meeting of the Institute of Food Technologists, June 16-20, Anaheim, CA, Book of Programs and Abstracts, #354, p.170.

Cadwallader, K.R. 1988. Bioconversion of (+)-limonene. 49th Annual Meeting of the Institute of Food Technologists, June 19-22, New Orleans, LA, Book of Programs and Abstracts, #293.

Students/Postdoctoral/Visiting Scholar Researchers Supervised (1991-1999)

Visiting Scientists
Yong-Jun Cha (LSU, MSU) – Chang Won National University
Hetty Voermans (LSU) – University of Wegeningen, The Netherlands

Postdoctoral Researchers
Hyung-Hee Baek (MSU)

Graduate Students Completed (LSU/MSU)

MS Students
Sun-Kyun Yoo (LSU)
Min Cai (MSU)
Hsin-Jen (Connie) Weng (MSU)
Shi-Guei (Scott) Fu (MSU)
Shin-Pei (Peggy) Yang (MSU)
Yi-Fang (Grace) Wu (MSU)
Leyla Yuksel (MSU)
JiHyun Jung (MSU)
Min-Sheng Su (MSU)
Gwo-Yuh Li (MSU)
Jeong Heo (MSU)

Ph.D. Students
Hyung-Hee Baek (LSU)
Hau Yin Chung (LSU)
Gyu-Hee (Chang) Lee (MSU)
Ranee Surakarnkul (MSU)
Yonca Karagul-Yuceer (MSU)

Students/Postdoctoral/Visiting Scholar Researchers Supervised (UIUC, 2000-present)

Visiting Scholars
Byung Hak Ahn (2006-2007) - Korea Food Research Institute, South Korea
Supeeraya Arsa (2013-2014) – Kasetsart University, Bangkok, Thailand
Mantana Buttara (2011-present) – Suranaree Univeristy of Technology, Thailand
Dong Chen (2015-present) – China Agricultural University, Beijing, China

Rongrong Chen (2016-present) - Ever Maple Food Science and Technology Co., Ltd., China
Arporn Jarunrattanasri (2003-2004) – Naresuan University, Thailand
Young-Suk Kim (2015-present) – Ewha Womans University
Patthamawadi Kiatbenjaku. (2009-2011, 2012) – Suranaree University of Technology, Thailand
Fareeya Kulapichitr (2016-present) – Chulalongkorn University, Thailand
Sasinee Kunyaboon (2016-present) - Suranaree University of Technology, Thailand
Nawaporn Lapsongphon (2010-2011) - Suranaree University of Technology, Thailand
Yuping Liu (2014-2015) – Beijing Technology and Business University, China
Yaowapa Lorjaroenphone (2006-2007) – Kasetsart University, Thailand
Xueli Pang (2013-2014) – China Agricultural University, Beijing, China
Sompoche Pojjanapimol (2003-2004) - Kasetsart University, Thailand
Primseri Rotsachakul (2004-2005) – Kasetsart University, Thailand
Huanlu Song (2005-2006) – Beijing Technology and Business University, Beijing, China
Patcharaporn Tinchan (2010-2012) – Kasetsart University, Thailand
Jihong Wu (2011-2012) – China Agricultural University, Beijing, China

Postdoctoral Researchers (UIUC)
Masud Huda (2002-2003)
Orasa Suriyaphan (2001, 2003)
Tanoj Singh (2001-2005)
Hun Kim (2005-2012)

Graduate Students Completed (UIUC)
Daniel Benitez (MS 2003)
Laura Brisske (MS 2003)
Erin Burnside (MS 2012)
Gabriela Carrasco (MS 2007)
Shondra Cook (MS 2007)
Edibe Erten (Ph.D. 2016)
Ming Chih Fang (Ph.D.2013)
Samantha Gardiner (MS 2015)
Elizabeth Genthner (MS 2010; Ph.D. 2014)
Theresa Gnadt (MS 2007)
Julia Gregg (Nonthesis MS, CHEFS Program, 2009)
Bethany Hausch (MS 2010)
Lauren Kelley (MS)
Sean LaFond (MS 2009)
Jacob Lahne (MS 2010)
Yaowapa Lorjaroenphone (Ph.D. 2012)
Patricio Lozano (MS 2005 & Ph.D. 2009)
Samantha McKenna (MS, 2016)
Joshua Moore (MS 2006)
Linda Moran (MS 2013)
Ana Carolina Mosca (MS 2007)
Jason Niermann (Nonthesis MS, CHEFS Program 2001)
Sirima Puangpraphant (MS 2007)
Bethany Richardson (MS 2013)
Inthawoot Suppavorasatit (Ph.D. 2012)
Chenxing Sun (3+2 program, Nonthesis MS)
Qiaoxuan Zhou (MS 2001 & Ph.D. 2005)

Current Graduate Students (2016)
Courtney Beckett (MS)
Chelsea Ickes (Ph.D.)
Fengjia Chen (MS, 3+1)
Bethany Hausch (Ph.D.)
Nagiat Hwasa (Ph.D.)
Jing (Jenny) Jin (MS, 3+1)
Tiandan Wu (MS)
Wenqi Zhu (Ph.D.)
Yun Yin (Ph.D.)

Undergraduate Student Researchers
Kelsey Choquette (2012-2013)
Diniene Chreaton, Rap II (2001)
Ashley Davis, Rap II (ACES) (2001) & UIUC Ronald E. McNair Summer Research Program (2004)
Yaozheng (Tess) Liu (2013-present)
Rosemary Nash (2011-2012)
Mary Nowak (2004-2005)
Jieun Oh (2013)
Abigail Pishevar (2014-2015)
David Potts (2008-2010)
Stephanie Sajuti (2005-2007)
Dylan Troth (2015-present)
Katrina Vlahovich (2001-2002)

Involvement in Professional Organizations (2000-present)

Leadership

Chair and Program Chair for the Division of Agricultural and Food Chemistry (AGFD) of the American Chemical Society, 2003-2005. Responsibilities include organizing AGFD technical programs for two ACS National Meetings (fall 2004 and spring 2005 meetings).

Editorial Board

Flavour and Frangrance Journal

Associate Editor

Journal of Food Science – *Food Chemistry*

Symposia Organized

Cadwallader, K.R. and Shahidi, F. 2017. ACS Symposium: *Chemistry of Clean Ingredients.* Washington, DC, August 20-24.

Cadwallader, K.R. and Chang, S.K.C. 2008. ACS Symposium: *Chemistry, Texture and Favor of Soy,* Philadelphia, PA, August 17-21.

Cadwallader, K.R. 2008. ACS Symposium: *Recent Developments in Flavor Chemistry*. 235th ACS National Meeting, New Orleans, LA, April 6-10.

Ho, C.-T., Weerasinghe, D.K. and Cadwallader, K.R. 2005. ACS Symposium: *Aroma-active Compounds in Foods and Reaction Favors: formation, measurement and stability.* 330th ACS National Meeting, Washington, DC, August 28 – September 1.

Cadwallader, K.R., P. Schieberle and M.J. Morello. 2005. ACS Symposium: *Grain-based Products: Health and Flavor, and Safety Aspects.* 330th ACS National Meeting, Washington, DC, August 28 – September 1.

Cadwallader, K.R. *Obesity Management: Role of Soy Foods.* University of Illinois, Champaign, IL, May 25-26, 2005.

Cadwallader, K.R., Drake, M.A. and McGorrin, R.J. 2004. ACS Symposium: *Chemistry and Flavor of Dairy Products.* 228th ACS National Meeting, Philadelphia, PA, August 22-26.

Weenen, H. and Cadwallader, K.R. (organizers). 2001. ACS Symposium: *Freshness and Shelf-life of Foods*. 222nd ACS National Meeting, Chicago, IL, August 26-30.

Cadwallader, K.R. and McGorrin, R.J. (organizers). 2001. IFT Symposium: *Developments in the Analysis of Complex Flavor Systems*. Annual Meeting of the Institute of Food Technologies, New Orleans, LA, June 23-27.

Rouseff, R.L. and Cadwallader, K.R. 1998. ACS Symposium: *Headspace Analysis of Foods and Flavors.* 214th ACS National Meeting, Boston, MA, August 23-27.

Shahidi, F. and Cadwallader, K.R. 1996. ACS Symposium: *Flavor and Lipid Chemistry of Seafoods.* 212th National Meeting of the American Chemical Society, Orlando, FL, August 25-29.

Workshops/Short courses (organized)

A Place for Soy in a Better School Lunch Program. University of Illinois, Urbana, IL, March 12, 2003.

Traditional and Emerging Methods for the Manufacture and Analysis of Food Flavorings. Faculty of Agriculture Industry, King Mongkut's Institute of Technology Ladkraband (KMITL), Bangkok, Thailand, July 29, 2002

Soy Flavor Workshop. ICSF Workshop (Klein, B.P. et al. coorganizers). Illinois Center for Soy Foods, University of Illinois, Urbana, IL, April, 2002/2003.

Workshops/Short courses (lecturer)

Nutraceuticals, Ingredients, Challenges, Formulations, Flavours and Health Effects. International Society for Nutraceutical and Functional Foods. Denpasar, Bali, Indonesia, October 11 – 12.

Flavor Chemistry. (course co-taught with Drs. Chi-Tang Ho and Soo-Yeun Lee), Taiwan National University, May 10-19, 2008.

Flavor Research Workshop. ACS Short Course (Acree, T.E., course director), American Chemical Society, Boston, MA, August 17-18, 2007.

Flavor Research Workshop. ACS Short Course (Acree, T.E. and Deibler, K., organizers), American Chemical Society, August 25-26, 2005.

Cadwallader, K.R. Flavoring soy products: masking and creating. Preconference AOAC Short Course. *Soyfoods: Ingredients, Preparation, and Utilization,* Koseoglu, S. and Shahidi, F. (organizers), Cincinnati, OH, May 9, 2005.

Cadwallader, K.R. 2004. Aroma and flavor aspects of soy beverages. Preconference Shortcourse: *Functional Beverages,* Koseoglu, S and Shahidi, F. (organizers), Worldnutra: International Conference and Exhibition on Nutraceutacals and Functional Foods, San Francisco, CA, Nov 8, 2004

Flavor Research Workshop. ACS Short Course (Acree, T.E., organizer), American Chemical Society, Boston, MA, August 16-17, 2002; New York, NY, September 7-11, 2003.

Cheese Flavor and Texture: Sensory and Instrumental Analysis. SDFRC Short Course (Drake, M.A., course director), Southeast Dairy Foods Research Center, North Carolina State University, Raleigh, NC, August 7-9, 2002.

Proposal Reviewer/Panel Participation (2010-present)

Panelist/Reviewer (2015) - Office of Scientific Quality Review (OSQR)-USDA-ARS, NP 306 Panel 14: Functional Foods (2015)

Panelist/Reviewer (2015) - Office of Scientific Quality Review (OSQR)-USDA-ARS, NP 107 Panel 14: Grain Engineering (2014)

Panelist/Reviewer (2014) - Office of Scientific Quality Review (OSQR)-USDA-ARS, NP 107 Panel 5: Food Chemistry Analysis (2014)

Panelist/Reviewer (2013) – 2012/2013 King Abdulaziz City for Science and Technology (KACST) Research Proposal Competition

EXHIBIT B

MATERIALS CONSIDERED

- U.S. Application Serial No. 14/797,006, Fraser, R. Brown, P.O., Karr, J. and Holz-Schietinger, C., 2014, Publication No. US 2015/0351435
- Hsieh, F-h and Huff, H.E. 2012. Meat analog compositions and process. U.S. Patent Application No. US 2012/0093994 ("the '994 application")
- Office Action Mailed June 6, 2016 and draft Response to same
- Declaration Under 37 CFR §1.132 of Celeste Holz-Schietinger, Ph.D.
- Calkins, C.R. and Hodgen, J.M. A fresh look at meat flavor. Meat Sci. 2007, 63-80.
- Proulx, A.K. and Reddy, M.B. Iron bioavailability of hemoglobin from soy root nodules using caco-2 cell culture model. J. Agric. Food Chem. 2006, 54, 1518-1522.
- Belitz, H.-D., Grosch, W., and Schieberle, P. Food Chemistry, 3rd revised edition. Springer-Verlag, Berlin (2006), p. 368.
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Attorney's Docket No.: 38767-0015001 / M3CON-Track 1

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

First Named Inventor	:	Rachel Fraser	Art Unit	: 1791
Serial No.	:	14/797,006	Examiner	: Elizabeth A. Gwartney
Filed	:	July 10, 2015	Conf. No.	: 8496
Title	:	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES		

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 37 CFR § 1.132

I, Devin Grant Peterson, PhD, hereby declare as follows:

1. I am currently a Professor in the Department of Food Science and Technology at the Ohio State University in Columbus, Ohio. I have held this position since early 2016. I have worked and taught in the field of Food Science since 2001. Prior to my current position, I held Professor/Associate Professor positions at the University of Minnesota in Saint Paul, Minnesota from 2009 to 2016; and was a faculty member in the Department of Food Science at the Pennsylvania State University in University Park, Pennsylvania from 2001 to 2009. In addition, I have been the Director of the Flavor Research and Education Center for the past seven years, at both the University of Minnesota and the Ohio State University.

2. I am a nationally recognized expert and scholar in the field of flavor science. In 2015, I received the Excellence in Flavor Science Award from the Flavor and Extract Manufacturers Association. In 2008, I received a Pan-American Nutrition, Food Science and Technology Award. In 2004, during my tenure at Pennsylvania State University, I was awarded the Presidential Early Career Award for Scientists and Engineers (PECASE), which was presented by the White House Office of Science and Technology Policy.

3. I received a Bachelor's of Science (B.S.) degree in Food Science in 1994 from the University of Manitoba in Winnipeg, Manitoba. I earned a Master's of Science (M.S.) degree in 1997 and a Doctor of Philosophy (Ph.D.) degree in 2001 in Flavor Chemistry from the Department of Food Science from the University of Minnesota in Saint Paul, Minnesota. I was named to the Dean's List during my academic career at both the University of Manitoba and at the University of Minnesota. While I was obtaining my Ph.D., I completed a six-month Internship as a Research Flavor Scientist at Givaudan Flavors in Cincinnati, Ohio.

4. A copy of my curriculum vitae is attached in **Exhibit A**.

5. My research focuses on food flavor and related chemistry, with an emphasis on identification of flavor stimuli (e.g., taste, aroma, chemesthetic, mouthfeel), pathways of formation and degradation of flavor stimuli, taste and aroma modulation, and mechanisms of delivery.

6. In preparing my Declaration, I have reviewed U.S. Patent Application No. 14/797,006 ("the '006 application"), as well as the Office Action mailed June 6, 2016, in which the claims of the '006 application were rejected as being anticipated by Proulx et al. (2006, J. Agric. Food Chem., 54:1518-22) ("Proulx") and as being obvious considering Hsieh et al. (US 2012/0093994) ("the '994 application") in view of Proulx. I also reviewed the Declaration under 37 CFR § 1.132 of Dr. Celeste Holz-Schietinger ("the Holz-Schietinger Declaration"). A list of the materials I reviewed in preparing this Declaration is provided in **Exhibit B**.

7. In forming my opinion, I reviewed and considered Applicants' experiments related to their discovery and demonstration that the addition of heme protein (in some cases as

leghemoglobin or LegH) surprisingly contributes to the development of meaty aroma in a "ground beef-like food product" comprising no animal products. These experiments are described in the '006 application and in the Holz-Schietinger Declaration.

8. Flavor is a complex sensation, consisting of different modalities such as olfaction (aroma), gustation (sweet, sour, salty, bitter, umami), and chemesthesis (heat, cooling, pain), as well as others. The aroma attributes are known to be a key component of flavor perception and have been the most widely studied by flavor chemists.

9. In my opinion, the scientific protocols in the '006 application and in the Holz-Schietinger Declaration for a) sample preparation and treatment; and b) chemical analysis of aqueous thermal reaction samples are reasonable and consistent with methods that would be used by skilled individuals in the art. Each of these aspects are reviewed in more detail below.

10. The Hotz-Schietinger Declaration describes chemical analysis of aqueous thermal reaction samples using art recognized instrumental aroma analysis methods, specifically HS-SPME combined with GC-MS, to monitor the effect of LegH addition on the formation of volatile compounds, specifically those with meaty aroma characteristics, during heating (i.e., cooking). The objective was to demonstrate that LegH was capable of promoting or catalyzing the formation of meaty aroma compounds via aqueous thermal reactions between ribose and cysteine. Three flavor reaction systems were compared, consisting of: 1) 1% (10 mg/mL) LegH alone (**LegH control**), 2) 50 mM ribose + 50 mM cysteine alone (**Rib + Cyst**) and 3) 1% (10 mg/mL) LegH + 50 mM ribose + 50 mM cysteine (**LegH + Rib + Cyst**). The above reaction mixtures were diluted in a buffer (50 mM potassium phosphate and 100 mM sodium chloride)

and adjusted to pH 6.5, which is near the physiological pH of ground beef. A 2 mL aliquot of each reaction mixture was transferred to sample vials (sealed) which were subsequently heated (150°C for 3 min) and then analyzed by HS-SPME-GC-MS.

11. The concentrations (i.e. 0.1 to 50 mM) of flavor precursors (i.e., ribose and cysteine) of the aqueous thermal reaction samples described in the '006 application and the Hotz-Schietinger Declaration are consistent with peer-reviewed publications that have used similar aqueous reaction samples to investigate the thermal generation of volatile aroma compounds contributing to the flavor sensation of a food product. While aqueous thermal reaction samples are simplistic compared to food matrices, they provide relevant guidance and generalized outcomes for understanding the influence of flavor precursors on the formation of volatile compounds contributing to aroma, and to the flavor sensation generally in a number of different food products.

12. For example, a widely utilized and referenced publication for understanding the impact of flavor precursor composition on the formation of volatile aroma compounds generated therefrom is Lane and Nursten, "Variety of odors produced in Maillard model systems and how they are influenced by reaction conditions" in Maillard Reaction in Foods and Nutrition, Waller and Feather, eds., ACS Symposium, Series No. 215, pp 141-158. This publication describes aqueous combinations of sugar and amino acid mixtures heated under different time and temperature conditions. Lane and Nursten provides directional guidance for flavor development, and how flavor is impacted by flavor precursors, in various types of food matrices.

13. It is my opinion that the experimental approach and subsequent data analysis techniques described in the Holz-Schietinger declaration are appropriate and acceptable for the purpose of evaluating and comparing aroma compound formation in the three aqueous thermal reaction systems under heating conditions. The aqueous thermal reaction samples described in the Holz-Schietinger Declaration are comparable with those described by Lane and Nursten. It is also my opinion that these conditions would simulate the cooking of a "ground beef-like food product" containing no animal products as described in the '006 application, and that the data obtained from the experimental approach would be reasonably predictive of the expected aroma compound formation from a "ground beef-like food product" containing no animal products and including the flavor reaction system components LegH, ribose, and cysteine.

14. It is well known that thermal generation of volatile aroma compounds in an aqueous reaction system is influenced by reaction temperature and pH. When utilizing aqueous reaction samples, the reaction systems often are sealed so they can be pressurized, which increases the temperature of the sample to simulate temperatures in excess of 100°C, which would be experienced with conventional food heating methods such as baking, grilling or pan-frying. The aqueous thermal reaction samples described in the '006 application and in the Holz-Schietinger Declaration were heated in sealed vials at 150°C, which would be consistent with temperatures experienced by common meat preparation (heating) techniques.

15. My research program has similarly utilized the benchtop autosampler apparatus utilized in the '006 application and the Holz-Schietinger Declaration to rapidly heat sealed vials of aqueous samples to between 100-150°C for the purpose of evaluating and comparing aroma

compound formation. See, for example, Kokkinidou and Peterson, 2013, Food Funct., 4:1093-104.

16. The slightly acidic pH value of 6 - 6.5 for the aqueous thermal reaction samples used in the '006 application and the Holz-Schietinger Declaration also are consistent with the typical intrinsic pH values of meat products, as well as with peer-reviewed publications evaluating flavor development, and how it is impacted by flavor precursors, in various types of food matrices, including in meat food products.

17. It is my opinion that the experimental approach and analytical methods used in the '006 application and the Holz-Schietinger Declaration for evaluating and comparing aroma compound formation in aqueous thermal reaction samples were consistent with those used by flavor scientists of ordinary skill in the art. For example, the headspace solid phase micro-extraction (SPME) sampling technique coupled with GCMS utilized in the '006 application and the Holz-Schietinger Declaration has been widely utilized for characterizing the aroma of foodstuffs, such as meat food products. See, for example, Elmore et al., 2001, "A comparison of headspace entrainment on Tenax with solid phase micro-extraction for the analysis of the aroma volatiles of cooked beef," in Headspace Analysis of Foods and Flavors; Rouseff and Cadwallader, eds, Springer. Indeed, more than 100 articles are reported in SciFinder when the key words "meat, aroma, SPME" are set as filters to screen the existing scientific literature.

18. It is well known that the aroma profiles of meat food products are impacted by sulfur-containing compounds (typically, reaction products of sulfur-containing amino acids) and lipid degradation/oxidation products that are generated during heating. The different chemical

classes of volatile compounds formed in the aqueous thermal reaction samples of the '006 application and the Holz-Schietinger Declaration are consistent with volatile aroma compounds reported in the literature evaluating volatile compound formation contributing to meat aroma and flavor sensation. See, for example, Elmore et al. (2002, J. Agric. Food Chem., 50:1126-32); Chen et al. (2003, "Influence of DNA on Volatile Generation from Maillard Reaction of Cysteine and Ribose," *in* Nutraceutical Beverages, ACS Symposium Series Vol 871); Mottram and Nobrega (2002, J. Agric. Food Chem., 50(14):4080-86); Schieberle and Hofmann ("Characterization of Key Odorants in Dry-Heated Cysteine—Carbohydrate Mixtures: Comparison with Aqueous Reaction Systems," *in* Flavor Analysis, ACS Symposium Series, Vol 705); Mottram and Mottram (2002, "An Overview of the Contribution of Sulfur-Containing Compounds to the Aroma in Heated Foods," *in* Heteroatomic Aroma Compounds, ACS Symposium Series, Vol 826); and Shu-C and Ho (1989, "Parameter Effects of the thermal reaction of cysteine and 2,5-dimethyl-4-hydroxy-3(2H)-furanone," *in* Thermal Generation of Aromas, ACS Symposium Series, Vol 409).

19. In my opinion, Applicant's experiments in the '006 application and the Holz-Schietinger Declaration demonstrate that the addition of LegH to an aqueous thermal reaction mixture comprising ribose and cysteine has the surprising result of enhancing the formation of volatile compounds associate with a meaty aroma. Significantly, the 2,851-fold increase in concentration of 2-acetylthiazole when LegH was combined with Ribose and Cysteine, which is shown in the Holz-Schietinger Declaration, in my opinion signifies that novel and unexpected chemical reactions occur in the presence of LegH. 2-acetylthiazole is particularly interesting as

it has been associated in the literature with a meaty/roasty aroma. See, for example, Chemistry and Technology of Flavours and Fragrances, 2005, Ed Rowe, Blackwell Publishing Ltd; Flavor of Meat and Meat Products, 1994, Ed Shahidi, Springer-Science+Business Media, BV; Handbook of Meat and Meat Processing, 2012, 2nd Ed, Ed Hui, CRC Press; Mottram and Mottram (2002, "An Overview of the Contribution of Sulfur-Containing Compounds to the Aroma in Heated Foods," *in* Heteroatomic Aroma Compounds, ACS Symposium Series, Vol 826); and Chen and Ho (2002, J. Agric. Food Chem., 50(8):2372–76).

20. Yield changes of 0.5- to 10-fold would be consistent with optimizing a reaction pathway, however, changes of greater than 1000-fold, as is seen with 2-acetylthiazole, indicate that new pathways of formation for meaty aromas result from the inclusion of LegH in the aqueous thermal reaction mixture.

21. Additionally, the Holz-Schietinger Declaration demonstrates that 2-methyl-3-furanthiol (MFT) and the major degradation product of MFT, the dimer, bis(2-methyl-3-furyl) disulfide, were detected only in the aqueous thermal reaction samples that contained Leghemoglobin. MFT is a very potent meaty odorant, with an extremely low odor threshold at 0.007 μg/kg in water, and is known to have an important contribution to meat aroma. See, for example, Kerscher and Grosch (1998, J. Agric. Food Chem., 46:1954-58); Tang et al. (2013, J. Sulfur Chem., 34:38-47); and Withcombe and Mussinan (1988, J. Food Sci., 53:658-60). It is surprising to me that an intense meaty aroma compound such as MFT was uniquely identified in the reaction mixture upon the addition of LegH.

22. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

Respectfully submitted,

Date: Dec 2, 2016

Devin Grant Peterson, Ph.D.

EXHIBIT A

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Devin G Peterson

Experience:

Aug/16 **Professor** **Food Science & Technology** **Director of the Flavor Research & Education Center**	The Ohio State University	Columbus, OH
June/12 – Aug/16 **Professor** **Food Science & Nutrition** **Director of the Flavor Research & Education Center**	The University of Minnesota	St. Paul, MN
Aug/09 – June/12 **Associate Professor** **Food Science & Nutrition** **Director of the Flavor Research & Education Center**	The University of Minnesota	St. Paul, MN
July/07 – Aug/09 **Associate Professor/Food Science**	The Pennsylvania State University	University Park, PA
March/01 – June/07 **Assistant Professor/Food Science**	The Pennsylvania State University	University Park, PA
Dec/99 – Feb./01 **Research Assistant**	University of Minnesota	St. Paul, MN
July/99 - Dec./99 **Research Flavor Scientist (Internship)**	Givaudan Flavors	Cincinnati, OH
Sept./94 - July/99 **Research Assistant**	University of Minnesota	St. Paul, MN
June/94 –Aug./94 **Information Technician**	The Canadian Wheat Board	Winnipeg, MB
May – Aug./92&93 **QC Technician**	Coca-Cola Bottling Company	Winnipeg, MB

Education:	1997 - 2001 **PhD/Food Science** (Dean's List)	University of Minnesota	St. Paul, MN
	1994 - 1997 **MS/Food Science** (Dean's List)	University of Minnesota	St. Paul, MN
	1990 – 1994 **BS/Food Science** (Dean's List)	University of Manitoba	Winnipeg, MB

Awards:	Flavor and Extract Manufacturers Association - Excellence in Flavor Science Award	2015
	Waters Centers of Innovation Award	2012
	Pan-American Nutrition, Food Science and Technology Award	2008
	Presidential Early Career Award for Scientist and Engineers (PECASE)	2004
	John Brandt Memorial Foundation Scholarship; National	2000
	M. E. Franks Scholarship; National	1998, 1999 & 2000
	IFT Grad. Fellowship–Soc. of Flavor Chemists Memorial; National	1998/99 & 2000/01
	AACC Graduate Fellowship; National	1997/98
	IFT Scholarship; Minnesota	1997 & 2000
	Triticus Award; Departmental	1996
	The Canadian Wheat Board Scholarship; National	1994

Peer Reviewed Publications (*italics* is corresponding author):

1. Raithore, S. and ***Peterson, D.G.*** 2016. Delivery of taste and aroma components in sugar-free chewing gum: mass balance analysis. Chemosensory Perception. *Accepted.*
2. Paravisini, L. and ***Peterson, D.G.*** 2016. Characterization of Browning Formation in Orange Juice during Storage. ACS Symposium Proceedings, Browned Flavors: Analysis, Formation, & Physiology. *Accepted.*
3. Kokkinidou, S. ***Peterson, D.G.*** 2016. Identification of compounds that contribute to trigeminal burn in aqueous ethanol solutions. Food Chem. 211: 757-762 http://www.sciencedirect.com/science/article/pii/S0308814616308068
4. Bin, Q. and ***Peterson, D.G.*** 2016. Identification of Bitter Compounds in Whole Wheat Bread Crumb. Food Chem. 203: 8–15, http://www.sciencedirect.com/science/article/pii/S0308814616301182
5. Yeo, L., Thompson, D. and ***Peterson, D.G.*** 2016. Inclusion Complexation of Flavor Compounds by Dispersed High-Amylose Maize Starch (HAMS) in an Aqueous Model System. Food Chem. *199*: 393–400. http://www.sciencedirect.com/science/article/pii/S0308814615303320
6. Yucel, U. and ***Peterson, D.G.*** 2015. Effect of Protein–Lipid–Salt Interactions on Sodium Availability in the Mouth and Consequent Perception of Saltiness: In Solutions. J. Agric. Food Chem., 63: 7487–7493. Impact Factor: 3.1 http://pubs.acs.org/doi/abs/10.1021/acs.jafc.5b02311
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10. Ronningen, I. and ***Peterson, D.G.*** 2015. Application of untargeted LC/MS techniques (Flavoromics) to identify changes related to freshness of food, *in* The Chemical Sensory Informatics of Food: Measurement, Analysis, Integration. Eds. Guthrie, et al.; ACS Symposium Series; American Chemical Society: Washington, DC, Vol. 1191, pg 269-277. http://pubs.acs.org/doi/pdf/10.1021/bk-2015-1191.ch019
11. Zhang, L., Xia, Y. and ***Peterson, D.G.*** 2014. Identification of bitter modulating Maillard-catechin reaction products. J. Agric. Food Chem. 62, 10092−10100. http://pubs.acs.org/doi/abs/10.1021/jf502040e
12. Karametsi, K., Kokkinidou, S., Ronningen, I. and ***Peterson, D.G.*** 2014. Identification of Bitter Peptides in Aged Cheddar Cheese. J. Agric. Food Chem. 62, 8034−8041. http://pubs.acs.org/doi/abs/10.1021/jf5020654
13. Troise, A. D. Fiore, A. Colantuono, A. Kokkinidou, S., **Peterson, D.G.** and *Fogliano, V.* 2014. Effect of olive mill wastewater phenol compounds on reactive carbonyl species and Maillard reaction end-products in ultrahigh-temperature-treated milk. J. Agric. Food Chem. 62, 10092−10100. http://pubs.acs.org/doi/abs/10.1021/jf501919y
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16. Kokkinidou, S. and ***Peterson, D.G.*** 2013. Response Surface Methodology as Optimization Strategy for Reduction of Reactive Carbonyl Species in Heat-processed Foods by Means of Phenolic Chemistry. Food and Function. 4(7): 1093-1104. http://pubs.rsc.org/en/content/articlelanding/2013/fo/c3fo60032g
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18. Moskowitz, M., Bing, Q., Elias, R. and ***Peterson, D.G.*** 2012. Influence of endogenous ferulic acid in whole wheat flour on bread crust aroma. J. Agric. Food Chem. 60: 11245-11252. http://pubs.acs.org/doi/abs/10.1021/jf303750y
19. Ferruzzi, M.G., **Peterson, D.G.**, Singh, R.P. Schwartz, S.J. and *Freedman, M.* 2012. Nutritional Translation Blended With Food Science: 21st Century Applications. Adv. Nutr. 3: 813–819. http://advances.nutrition.org/content/3/6/813
20. Bin, Q., Jiang, D. Cho, I.H., ***Peterson, D. G.*** 2012. Chemical markers for bitterness in wheat bread. 2012. Flavour Fragr. J. 27, 454–458. http://onlinelibrary.wiley.com/doi/10.1002/ffj.3124/abstract

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22. Bin, Q. **Peterson, D.G.** and *Elias, R.J.* 2012. Influence of phenolic compounds on the mechanisms of pyrazinium radical generation in the Maillard reaction. J. Agric. Food Chem. *60* (21), pp 5482–5490. http://pubs.acs.org/doi/full/10.1021/jf3010838
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24. Moskowitz, M. and **Peterson, D. G.** 2011. Hydroxycinnamic acid-Maillard reactions in simple aqueous model systems. *in* Controlling Maillard Pathways. Washington, D.C.: American Chemical Society Symposium Proceedings 1042, pp 53-62. http://pubs.acs.org/doi/abs/10.1021/bk-2010-1042.ch006
25. Holt, A., Potineni, R. and **Peterson, D.G.** 2011. Flavor ingredient interactions in confections. Accepted. 12th Weurman Symposium Proceedings, Interlaken, Switzerland. *Pg 139-142.* https://home.zhaw.ch/~vere/pdf/Teil37 - Expression of Multidisciplinary.pdf
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Patents (inventor):
International Application (PCT). April 21st, 2015. Peterson, D. and Kokkinidou, S. Methods of Making Modified Alcohol Containing Products.

USA application. An umami-enhanced food product and method of enhancing the umami taste of food products.

Courses Taught:
Flavor Chemistry (FScN 8337), Flavor Technology (FScN 5521); Current Issues in Food Science (FScN 8318); Food Chemistry and Functional Foods (FScN 4112)

Professional Memberships:
American Chemical Society
Institute of Food Technologists

EXHIBIT B

MATERIALS CONSIDERED

- U.S. Application Serial No. 14/797,006, Fraser, R. Brown, P.O., Karr, J. and Holz-Schietinger, C., 2014, Publication No. US 2015/0351435
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- Spence, C. 2015. Multisensory flavor perception. Cell 161: 24-35.

EXHIBIT A

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Experience:

Aug/16 **Professor** **Food Science & Technology** **Director of the Flavor Research & Education Center**	The Ohio State University	Columbus, OH
June/12 – Aug/16 **Professor** **Food Science & Nutrition** **Director of the Flavor Research & Education Center**	The University of Minnesota	St. Paul, MN
Aug/09 – June/12 **Associate Professor** **Food Science & Nutrition** **Director of the Flavor Research & Education Center**	The University of Minnesota	St. Paul, MN
July/07 – Aug/09 **Associate Professor/Food Science**	The Pennsylvania State University	University Park, PA
March/01 – June/07 **Assistant Professor/Food Science**	The Pennsylvania State University	University Park, PA
Dec/99 – Feb./01 **Research Assistant**	University of Minnesota	St. Paul, MN
July/99 - Dec./99 **Research Flavor Scientist (Internship)**	Givaudan Flavors	Cincinnati, OH
Sept./94 - July/99 **Research Assistant**	University of Minnesota	St. Paul, MN
June/94 –Aug./94 **Information Technician**	The Canadian Wheat Board	Winnipeg, MB
May – Aug./92&93 **QC Technician**	Coca-Cola Bottling Company	Winnipeg, MB

Education:	1997 - 2001 **PhD/Food Science** (Dean's List)	University of Minnesota	St. Paul, MN
	1994 - 1997 **MS/Food Science** (Dean's List)	University of Minnesota	St. Paul, MN
	1990 – 1994 **BS/Food Science** (Dean's List)	University of Manitoba	Winnipeg, MB

Awards:	Flavor and Extract Manufacturers Association - Excellence in Flavor Science Award	2015
	Waters Centers of Innovation Award	2012
	Pan-American Nutrition, Food Science and Technology Award	2008
	Presidential Early Career Award for Scientist and Engineers (PECASE)	2004
	John Brandt Memorial Foundation Scholarship; National	2000
	M. E. Franks Scholarship; National	1998, 1999 & 2000
	IFT Grad. Fellowship–Soc. of Flavor Chemists Memorial; National	1998/99 & 2000/01
	AACC Graduate Fellowship; National	1997/98
	IFT Scholarship; Minnesota	1997 & 2000
	Triticus Award; Departmental	1996
	The Canadian Wheat Board Scholarship; National	1994

Peer Reviewed Publications (*italics* is corresponding author):

1. Raithore, S. and ***Peterson, D.G.*** 2016. Delivery of taste and aroma components in sugar-free chewing gum: mass balance analysis. Chemosensory Perception. *Accepted.*
2. Paravisini, L. and ***Peterson, D.G.*** 2016. Characterization of Browning Formation in Orange Juice during Storage. ACS Symposium Proceedings, Browned Flavors: Analysis, Formation, & Physiology. *Accepted.*
3. Kokkinidou, S. ***Peterson, D.G.*** 2016. Identification of compounds that contribute to trigeminal burn in aqueous ethanol solutions. Food Chem. 211: 757-762 http://www.sciencedirect.com/science/article/pii/S0308814616308068
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11. Zhang, L., Xia, Y. and ***Peterson, D.G.*** 2014. Identification of bitter modulating Maillard-catechin reaction products. J. Agric. Food Chem. 62, 10092−10100. http://pubs.acs.org/doi/abs/10.1021/jf502040e
12. Karametsi, K., Kokkinidou, S., Ronningen, I. and ***Peterson, D.G.*** 2014. Identification of Bitter Peptides in Aged Cheddar Cheese. J. Agric. Food Chem. 62, 8034−8041. http://pubs.acs.org/doi/abs/10.1021/jf5020654
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http://www.sciencedirect.com/science/article/pii/S0032959203004618
51. **Peterson, D.G.** and V.A. Totlani. 2005. Influence of flavonoids on the thermal generation of aroma compounds. in Phenolics in Foods and Natural Health Products, New York, ACS Symposium Series #909, September 7-11, 2003, pg 143-160.
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52. Schober, A.L. and ***Peterson, D.G.*** 2004. Flavor release and perception in hard candy: Influence of flavor compound-compound interactions. J. Ag. Fd. Chem. 52: 2623-2627.
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http://pubs.acs.org/doi/full/10.1021/jf0354287
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Patents (inventor):
International Application (PCT). April 21st, 2015. Peterson, D. and Kokkinidou, S. Methods of Making Modified Alcohol Containing Products.

USA application. An umami-enhanced food product and method of enhancing the umami taste of food products.

Courses Taught:
Flavor Chemistry (FScN 8337), Flavor Technology (FScN 5521); Current Issues in Food Science (FScN 8318); Food Chemistry and Functional Foods (FScN 4112)

Professional Memberships:
American Chemical Society
Institute of Food Technologists

EXHIBIT B

MATERIALS CONSIDERED

- U.S. Patent Application No. 14/797,006 ("the '006 application")
- Office Action mailed June 6, 2016
- Proulx et al. (2006, J. Agric. Food Chem., 54:1518-22) ("Proulx")
- Hsieh et al. (US 2012/0093994) ("the '994 application")
- Declaration under 37 CFR § 1.132 of Dr. Celeste Holz-Schietinger ("the Holz-Schietinger Declaration")
- Lane and Nursten, "Variety of odors produced in Maillard model systems and how they are influenced by reaction conditions" in Maillard Reaction in Foods and Nutrition, Waller and Feather, eds., ACS Symposium Series No. 215, pp 141-158
- Kokkinidou and Peterson, 2013, Food Funct., 4:1093-104
- Elmore et al., 2001, "A comparison of headspace entrainment on Tenax with solid phase micro-extraction for the analysis of the aroma volatiles of cooked beef," in Headspace Analysis of Foods and Flavors; Rouseff and Cadwallader, eds, Springer
- Elmore et al. (2002, J. Agric. Food Chem., 50:1126-32)
- Chen et al. (2003, "Influence of DNA on Volatile Generation from Maillard Reaction of Cysteine and Ribose," *in* Nutraceutical Beverages, ACS Symposium Series Vol 871)
- Mottram and Nobrega (2002, J. Agric. Food Chem., 50(14):4080–86)
- Schieberle and Hofmann ("Characterization of Key Odorants in Dry-Heated Cysteine—Carbohydrate Mixtures: Comparison with Aqueous Reaction Systems," *in* Flavor Analysis, ACS Symposium Series, Vol 705)

- Mottram and Mottram (2002, "An Overview of the Contribution of Sulfur-Containing Compounds to the Aroma in Heated Foods," *in* Heteroatomic Aroma Compounds, ACS Symposium Series, Vol 826)
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- Chemistry and Technology of Flavours and Fragrances, 2005, Ed David Rowe, Blackwell Publishing Ltd
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- Kerscher and Grosch (1998, J. Agric. Food Chem., 46:1954-58)
- Tang et al. (2013, J. Sulfur Chem., 34:38-47)
- Withcombe and Mussinan (1988, J. Food Sci., 53:658-60)

DECLARATION UNDER 37 CFR §1.132

I, Celeste Holz-Schietinger, Ph.D., hereby declare as follows:

1. I am currently a Principal Scientist at Impossible Foods in Redwood City, California. I have held this position since 2015, and I am responsible for leading the Flavor Department at Impossible Foods. In this role, I direct flavor technology research as well as flavor product integration. I began working for Impossible Foods in 2012 as a Scientist, and I was elevated to a Senior Scientist in 2013. While at Impossible Foods, I have been the lead chemist on the identification, characterization and manipulation of biological flavor catalysis and potentiation, including through the use of heme-containing proteins.

2. I received my Bachelors of Science (B.S.) degree in Molecular Biology and Evolution, with a minor in Chemistry, in 2005 from The Evergreen State College in Olympia, Washington, and I received my Doctor of Philosophy (Ph.D.) degree in Biochemistry from the University of California at Santa Barbara in 2012. I was an Enzymology Research Associate at the University of California at Santa Barbara from 2007 to 2012, and I also was a Biochemistry Visiting Associate at Stanford University in 2009.

3. A copy of my curriculum vitae is attached as **Exhibit A**.

4. Gas Chromatography-Mass Spectroscopy (GCMS) and Gas Chromatography-Olfactory (GCO) methods were used to evaluate the production of volatile compounds during cooking of flavor reaction mixtures, and in particular, to evaluate the production of volatile compounds that are associated with meaty aroma. The first flavor reaction mixture set contained only Leghemoglobin (LegH-only) at different concentrations (0.01%, 0.2%, 1%, and 3%) (note 1% =10 mg/mL); the second set contained a mixture representing the estimated concentrations of amino acids, sugars, and other small molecules in beef based on their values reported in literature as shown below (Table 1, "the Precursor Mix"):

Table 1

Precursor Mix	
Chemical entity	**mM**
Alanine	5.6
Arginine	0.6
Asparagine	0.8
Aspartate	0.8
Cysteine	0.8

Glutamic acid	3.4
Glutamine	0.7
Glycine	1.3
Histidine	0.6
Isoleucine	0.8
Leucine	0.8
Lysine	0.7
Methionine	0.7
Phenylalanine	0.6
Proline	0.9
Threonine	0.8
Tryptophan	0.5
Tyrosine	0.6
Valine	0.9
Glucose	5.6
Ribose	6.7
Maltodextrin	5
Thiamine	0.5
GMP	0.24
IMP	0.6
Lactic acid	1
Creatine	1
NaCl	10
KCl	10
Potassium phosphate pH 6.5	10

and the third contained both Leghemoglobin at different concentrations (0.01%, 0.05% 0.2%, 0.5%, 1%, and 3%) and the Precursor Mix. All flavor reaction mixtures were adjusted to pH 6.5 using 50 mM of potassium phosphate buffer and 100 mM of sodium chloride buffer. Two mL of each flavor reaction mixture were prepared in 10 mL GCMS glass vials (referred to as "flavor reaction mixture samples" or "samples" hereinafter). Each sample was analyzed in duplicate.

5. An Agilent 7890A GC system coupled with a Leco TruTOF HT system was used along with a Gerstel MultiPurpose Sampler for auto-sampling. The Gerstel auto-sample was used to cook, cool, and perform HS-SPME on each of the samples. Each sample was heated to 150°C for 3 minutes with 750 rpm of agitation to "cook" the material. This was followed by a two-phase active cooling step that decreased the agitator temperature from 150°C to 50°C. The sample was extracted for 20 minutes at 50°C with 500 rpm of agitation using a 50/30 µm

Divinylbenzene / Carboxen / Polydimethylsiloxane (DVB/CAR/PDMS), Stable flex, 23 Ga, Autosampler (Supleco, Cat# 57298-U) fiber.

6. Extracted samples were run on the GCMS by desorbing the SPME fiber in the Gerstel controled CIS inlet for 6 minutes at 240°C. The desorbed sample was separated on a 60-meter wax column (Agilent, VF-WAXms 60 m x 0.25 mm x 0.25 mm, Part# CP9207) using a 35 minutes GCMS method (35°C for 2 minutes, ramp of 5C°/min until 172°C, ramp of 60C°/min until 255°C, 255°C for 4 minutes) and 1.75 mL/min Helium flow rate and splitless mode. Separated compounds were analyzed by the mass spectrometer, and all data was collected over a 14-500 μ mass range, with an acquisition rate of 10 spectra/sec, and a detector voltage off-set of 100.

7. Samples were then analyzed using Leco ChromaTOF optimized for TruTOF HT (Version 4.51.6.0) coupled with the NIST MS Search 2.2. The identity of each peak in each sample was identified in a two-step process. First, the mass spectrum of each peak with a signal to noise greater than 50 was matched to a mass spectrum in the NIST library using a match threshold of 650. Additionally, an internally developed calibration method was applied to the data set to confirm the identity of the compounds of interest. In the second step, the Statistical Compare function of ChromaTOF was used to align the named analytes across all samples in a set. The criteria for aligning a single peak across all samples were a similarity score (similarity between the peak spectra across all samples) of 700, a maximum retention time difference of 3 seconds, and the peak must have been present in at least two samples.

8. The amount of each aroma compound was determined by averaging the signal intensities from the duplicate experiments. The amount of each aroma compound that can be attributed to leghemoglobin (LegH)[1] was determined by using the average signal intensity for each aroma compound produced in the LegH-only sample as a baseline to standardize the average signal intensity for the same aroma compound produced in the LegH + Precursor Mix sample. The fold-change in each aroma compound was determined by dividing the signal intensity produced in the presence of LegH (i.e., in the LegH + Precursor Mix sample) by the signal intensity produced in the absence of LegH (i.e., Precursor Mix sample). If the signal intensity in the absence of LegH (i.e., Precursor Mix) was not detectable, the aroma compound

[1] Aroma compounds that can be attributed to LegH refer to compounds that are, independently, either not detectably produced or produced in a detectably smaller amount upon cooking compared with the control samples (e.g., LegH-alone or Precursor Mix in the absence of LegH).

produced in the presence of LegH (i.e., in the LegH + Precursor Mix sample) was indicated as "novel," meaning that the aroma compound was newly produced (to the limits of detection of the technique) in the LegH + Precursor Mix sample.

9. The results of the above-described experiments are shown in **Exhibit B**. The first column shows the list of aroma compounds, grouped into eight (8) different classes of chemicals. The next columns show the amount of each aroma compound, calculated as described above, for each of the samples (LegH only, Precursor Mix only, and LegH + Precursor Mix) followed by a column indicating the fold-change, calculated as described above, or indicating that the aroma compound is novel as described above. The next column provides, if available, one or more peer-reviewed publications in which that particular aroma compound is associated with meaty aroma.

10. The presence of **0.01% LegH** with precursor mix resulted in the production of **twenty-two (22)** novel meat associated aroma compounds upon cooking that were not produced in the absence of the LegH. The 22 different novel aroma compounds can be divided into eight (8) different classes of compounds as follows. Representative aroma compounds in each class are shown below; the full list can be found in Exhibit B.

- *Lactones*: one (1) novel lactone was produced (i.e., dihydro-5-pentyl-2(3H)-furanone);
- *Oxygen (O)- and/or Nitrogen (N)-containing heterocyclic*: no (0) novel O- and/or N-containing heterocyclic compounds were produced;
- *Sulfur (S)-containing heterocyclic*: five (5) novel S-containing heterocyclic compounds were produced (i.e., 3-thiophenemethanol, 3-methyl-thiophene, 2-ethyl-thiophene, 2-methyl-5-propyl-thiophene, 4-(methylthio)thiophenol);
- *Carboxylic acids*: three (3) novel carboxylic acids were produced (i.e., hexanoic acid, octanoic acid, propanoic acid);
- *Aldehydes*: eight (8) novel aldehydes were produced (i.e., decanal, undecanal, butanal, heptanal, 2-methyl-propanal, propanal, 3-methyl-butanal, 3-methyl-2-butenal);
- *Sulfides*: two (2) novel sulfide compounds were produced (i.e., dimethyl sulfide, methanethiol);

- *Ketones*: three (3) novel ketones were produced (i.e., 2-nonanone, 2-heptanone, 2,3-pentanedione); and
- *Alcohols*: no (0) novel alcohols were produced.

11. The presence of **0.2% LegH** with precursor mix resulted in the production of **twenty-seven (27)** novel meat associated aroma compounds upon cooking that were not produced in the absence of the LegH. The 27 different novel aroma compounds can be divided into eight (8) different classes of compounds as follows. Representative aroma compounds in each class are shown below; the full list can be found in Exhibit B.

- *Lactones*: no (0) novel lactones were produced;
- *Oxygen (O)- and/or Nitrogen (N)-containing heterocyclic*: two (2) novel O- and/or N-containing heterocyclic compound was produced (i.e., 2,5-dimethyl-pyrazine, indole);
- *Sulfur (S)-containing heterocyclic*: five (5) novel S-containing heterocyclic compounds were produced (i.e., 3-thiophenemethanol, 3-methyl-thiophene, 2-ethyl-thiophene, 2-methyl-5-propyl-thiophene, 4-(methylthio)thiophenol);
- *Carboxylic acids*: four (4) novel carboxylic acids were produced (i.e., hexanoic acid, hexadecanoic acid, octanoic acid, propanoic acid);
- *Aldehydes*: nine (9) novel aldehydes were produced (i.e., decanal, undecanal, butanal, heptanal, 2-methyl-propanal, propanal, hexanal, 3-methyl-butanal, 3-methyl-2-butenal);
- *Sulfides*: four (4) novel sulfide compounds were produced (i.e., dimethyl sulfide, dimethyl trisulfide, dimethyl disulfide, methanethiol);
- *Ketones*: three (3) novel ketones were produced (i.e., 2-nonanone, 2-heptanone, 2,3-pentanedione); and
- *Alcohols*: no (0) novel alcohols were produced.

12. The presence of **1% LegH** with precursor mix resulted in the production of **thirty two (32)** novel meat associated aroma compounds upon cooking that were not produced in the absence of the LegH. The 32 different novel aroma compounds can be divided into eight (8) different classes of compounds as follows. Representative aroma compounds in each class are shown below; the full list can be found in Exhibit B.

- *Lactones*: no (0) novel lactones were produced;

- *Oxygen (O)- and/or Nitrogen (N)-containing heterocyclic*: four (4) novel O- and/or N-containing heterocyclic compounds were produced (i.e., 2-pentyl-pyridine, 2-ethyl-5-methyl- pyrazine, 2,5-dimethyl-pyrazine, indole);
- *Sulfur (S)-containing heterocyclic*: seven (7) novel S-containing heterocyclic compounds were produced (i.e., 3-thiophenemethanol, 1-(4,5-dihydro-2-thiazolyl)-ethanone, 3-methyl-thiophene, 2-ethyl-thiophene, 2-methyl-5-propyl-thiophene, 4-(methylthio)thiophenol, 2,4-dimethyl-thiazole);
- *Carboxylic acids*: four (4) novel carboxylic acids were produced (i.e., hexanoic acid, hexadecanoic acid, octanoic acid, propanoic acid);
- *Aldehydes*: ten (10) novel aldehydes were produced (i.e., decanal, undecanal, butanal, heptanal, 2-methyl-propanal, propanal, hexanal, 3-methyl-butanal, pentanal, 3-methyl-2-butenal);
- *Sulfides*: three (3) novel sulfide compounds were produced (i.e., dimethyl sulfide, dimethyl trisulfide, dimethyl disulfide);
- *Ketones*: three (3) novel ketones were produced (i.e., 2-nonanone, 2-heptanone, 2,3-pentanedione); and
- *Alcohols*: one (1) novel alcohol was produced (i.e., furaneol).

13. The presence of **3% LegH** with precursor mix resulted in the production of **thirty-three (33)** novel meat associated aroma compounds upon cooking that were not produced in the absence of the LegH. The 33 different novel aroma compounds can be divided into eight (8) different classes of compounds as follows. Representative aroma compounds in each class are shown below; the full list can be found in Exhibit B.

- *Lactones*: one (1) novel lactone was produced (i.e., 5-ethyldihydro-2(3H)-furanone);
- *Oxygen (O)- and/or Nitrogen (N)-containing heterocyclic*: four (4) novel O- and/or N-containing heterocyclic compounds were produced (i.e., 2-pentyl-pyridine, 2-ethyl-5-methyl- pyrazine, 2,5-dimethyl-pyrazine, indole);
- *Sulfur (S)-containing heterocyclic*: seven (7) novel S-containing heterocyclic compounds were produced (i.e., 3-thiophenemethanol, 1-(4,5-dihydro-2-thiazolyl)-ethanone, 3-methyl-thiophene, 2-ethyl-thiophene, 2-methyl-5-propyl-thiophene, 4-(methylthio)thiophenol, 2,4-dimethyl-thiazole);

- *Carboxylic acids*: four (4) novel carboxylic acids were produced (i.e., hexanoic acid, hexadecanoic acid, octanoic acid, propanoic acid);
- *Aldehydes*: ten (10) novel aldehydes were produced (i.e., decanal, undecanal, butanal, heptanal, 2-methyl-propanal, propanal, hexanal, 3-methyl-butanal, pentanal, 3-methyl-2-butenal);
- *Sulfides*: three (4) novel sulfide compound was produced (i.e., dimethyl sulfide, dimethyl trisulfide, dimethyl disulfide);
- *Ketones*: three (3) novel ketones were produced (i.e., 2-nonanone, 2-heptanone, 2,3-pentanedione); and
- *Alcohols*: one (1) novel alcohol was produced (i.e., furaneol).

14. Across the four concentration ranges of LegH, **35** different novel aroma compounds were identified, each of which have been associated, directly or indirectly, with meaty aroma; see, for example, the references provided in **Exhibit B**. Specifically,

- twenty (20) of those novel compounds were produced in all four LegH reactions (0.01%, 0.2%, 1% and 3%) (i.e., 3-thiophenemethanol, 3-methyl-thiophene, 2-ethyl-thiophene, 2-methyl-5-propyl-thiophene, 4-(methylthio)thiophenol, hexanoic acid, octanoic acid, propanoic acid, decanal, undecanal, butanal, heptanal, 2-methyl-propanal, propanal, 3-methyl-butanal, 3-methyl-2-butenal, dimethyl sulfide, 2-nonanone, 2-heptanone, and 2,3-pentanedione);
- six (6) of those novel compounds were produced in all LegH reactions except at the lowest concentration (i.e., 0.2%, 1% and 3% LegH, but not 0.01% LegH) (i.e., 2,5-dimethyl-pyrazine, hexadecanoic acid, hexanal, dimethyl trisulfide, dimethyl disulfide, and indole); and
- four (4) of those novel compounds were produced in the presence of the higher concentrations of LegH (i.e., 1% and 3% LegH, but not 0.01% or 0.2% LegH) (i.e., 1-(4,5-dihydro-2-thiazolyl)-ethanone, 2,4-dimethyl-thiazole, pentanal, and furaneol).

15. The presence of LegH in the flavor reaction mixture with the Precursor Mix also resulted in a synergistic and dramatic increase in the amount of numerous meat associated aroma compounds produced upon heating compared to the amount of the same aroma compounds

produced upon heating in the absence of LegH (i.e., in the Precursor Mix sample) or in the LegH sample. For example, in the presence of LegH, the amount of:

- 3-thiophenecarboxaldehyde was increased almost 10-fold in the presence of 0.01% LegH, more than 250-fold in the presence of 0.2% LegH, about 3600-fold in the presence of 1% LegH, and over 4300-fold in the presence of 3% LegH;
- methyl-2-thiophenecarboxaldehyde was increased over 350-fold in the presence of 1% LegH and over 1460-fold in the presence of 3% LegH.;
- methyl pyrazine was increased over 10-fold in the presence of 0.2% LegH, over 120-fold in the presence of 1% LegH, and almost 150-fold in the presence of 3% LegH;
- 2-methyl-3-furanthiol was increased over 120-fold in the presence of 0.01% LegH, and about 320-fold in the presence of 0.2% LegH;
- dihydro-3-(2H)-thiophenone was increased almost 5-fold in the presence of 0.2% LegH, over 15-fold in the presence of 1% LegH, and over 30-fold in the presence of 3% LegH; and
- 2,3-butanedione was increased over 8-fold in the presence of 0.2% LegH, almost 650-fold in the presence of 1% LegH, and almost 620-fold in the presence of 3% LegH.
- 2-butanone was increased over 15-fold in the presence of 0.2% LegH, over 125-fold in the presence of 1% LegH, and over 165-fold in the presence of 3% LegH.
- 2-acetylthiazole was increased over 8-fold in the presence of 0.2% LegH, almost 215-fold in the presence of 1% LegH, and almost 275-fold in the presence of 3% LegH.
- furfural was increased almost 40-fold in the presence of 1% LegH and over 30-fold in the presence of 3% LegH.

Significantly, each of these compounds have been associated, either directly or indirectly, with meaty or beefy aroma. See, for example, the references cited in **Exhibit B**. Specifically, 2-

methyl-3-furanthiol[2,3] and 2-acetylthiazole[4], for example, are considered important components in the aroma of meat or beef.

[2] See paragraph 15 in the Declaration under 37 CFR § 1.132 by Dr. Keith R. Cadwallader ("the Cadwallader Declaration"), submitted in U.S. Application No. 14/797,006, now U.S. Patent No. 9,700,067, on December 5, 2016, which states:

> Among the 28 compounds identified in [the accompanying Holz-Schietinger Declaration] experiments, two in particular are well-known to possess meaty sensory characteristics and to be important and potent odorants in cooked meat. Specifically, 2-methyl-3-furanthiol (MFT) and its dimerized form bis(2-methyl-3-furyl)disulfide (MFT-MFT) are important aroma compounds in cooked meat (Tang et al., 2013). Both compounds have extremely low odor detection thresholds of 0.7 parts-per-trillion (pptr) and 0.02 pptr, respectively (Belitz et al. 2006), and are usually only present in low abundance (pptr range) in cooked meat (Tang et al., 2013). Despite their low abundance in meat they are still among the most impactful aroma compounds because of their extremely low odor detection thresholds.

[3] See paragraph 21 in the Declaration under 37 CFR § 1.132 by Dr. Devin Grant Peterson ("the Peterson Declaration"), submitted in U.S. Application No. 14/797,006, now U.S. Patent No. 9,700,067, on December 5, 2016, which states:

> Additionally, the [accompanying Holz-Schietinger Declaration] demonstrates that 2-methyl-3-furanthiol (MFT) and the major degradation product of MFT, the dimer, bis(2-methyl-3-furyl) disulfide, were detected only in the aqueous thermal reaction samples that contained Leghemoglobin. MFT is a very potent meaty odorant, with an extremely low odor threshold at 0.007 µg/kg in water, and is known to have an important contribution to meat aroma. See, for example, Kerscher and Grosch (1998, J. Agric. Food Chem., 46:1954-58); Tang et al. (2013, J. Sulfur Chem., 34:38-47); and Withcombe and Mussinan (1988, J. Food Sci., 53:658-60). It is surprising to me that an intense meaty aroma compound such as MFT was uniquely identified in the reaction mixture upon the addition of LegH.

[4] See paragraph 19 of the Peterson Declaration, submitted in U.S. Application No. 14/797,006, now U.S. Patent No. 9,700,067, on December 5, 2016, which states:

> 2-acetylthiazole is particularly interesting as it has been associated in the literature with a meaty/roasty aroma. See, for example, Chemistry and Technology of Flavours and Fragrances, 2005, Ed Rowe, Blackwell Publishing Ltd; Flavor of Meat and Meat Products, 1994, Ed Shahidi, Springer-Science+Business Media, BV; Handbook of Meat and Meat Processing, 2012, 2nd Ed, Ed Hui, CRC Press; Mottram and Mottram (2002, "An Overview of the Contribution of Sulfur-Containing Compounds to the Aroma in Heated Foods," in Heteroatomic Aroma Compounds, ACS Symposium Series, Vol 826); and Chen and Ho (2002, J. Agric. Food Chem., 50(8):2372–76).

16. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

Respectfully submitted,

Date: 8/17/17

Celeste Holz-Schietinger

EXHIBIT A

Celeste Holz-Schietinger, PhD

Principal Scientist, Impossible Foods

Redwood City, CA
celeste.holz@impossiblefoods.com

Innovative R&D Scientist and Product Developer responsible for leading the Flavor Department at Impossible Foods. Technical skills include enzymology, flavor chemistry, sensory science, lipid and protein biochemistry, protein purification, analytical chemistry, metabolic pathways and small molecule identification.

Education

Ph.D. Biochemistry, **University of California** at Santa Barbara — *2007-2012*
from the Biomolecular Science and Engineering Program

B.S. Molecular Biology & Evolution, minor in Chemistry, — *2001- 2005*
The Evergreen State College, Olympia, WA

Research

Principal Scientist, *Impossible Foods, Dr. Pat Brown* — *2015- Current*

Senior Scientist, *Impossible Foods, Dr. Peg Scott, Dr. Chris Davis* — *2013- 2015*

Scientist, *Impossible Foods, Dr. Chris Davis* — *2012-2013*

- R&D scientist and flavor department leader at Impossible Foods, directing flavor technology research and flavor product integration.
- Manage a team of scientists to understand the fundamental biology and chemistry of flavor perception and flavor creation. We identify the catalysis and precursors that create aroma compounds, determine how aroma compounds create particular flavors profiles, and then how to recreate these flavors in products.
- Lead chemist on the identification, characterization and manipulation of biological flavor catalysis, including heme-proteins, microbes, and enzymes.
- Support off flavor identification and removal in products and feedstock with product developers, protein biochemist and process engineers.
- Flavor product integration, leading efforts for creating meat replicas from plants that taste and smell like animal based meats, working closely with manufacturing to scale the process, and marketing to meet consumers desires.
- Previous roles also included but not limited to sensory panel leader, lipid group leader, and product developer creating nut-base cheese (Kite Hill Cheese).

Enzymology Research Associate, *UC Santa Barbara, Dr. Norbert Reich* — *2007 –2012*

- Researched the underlying chemical mechanisms that regulate how the genetic code is translated to creates phenotypic variation in human cells; particularly, understanding the developmental enzymes (DNMT3B and DNMT3A) and their modulators.
- Identified the mechanisms and structural changes that occurred with mutated forms of DNMT3A, highly associated with diseased states that change gene expression.

- Particular specialty was the development and execution of kinetic and biophysical assays to characterize the enzymology and regulation of the *de novo* DNA methyltransferase (DNMT3A).
- Mentored and lead 6 undergraduate researchers.

Biochemistry Visiting Associate, *Stanford University, Dr. Pat Brown (collaboration 2008-2010) 06-11/2009*

- Global identification of RNA associated with developmental enzymes (DNMT3B and DNMT3A) through cellular differentiation and correlated to gene expression and DNA methylation patterns. Developed RNA library construction protocols for next generation sequencing, and became skilled in microarray, hES cell culture, and fluorescence microscopy.

Scientist in Endocrinology, *U.S. Geological Survey* *05-09/2006*

- Conducted research on the stress response of translocation on the desert tortoise, though ELISA-based assays, leading team of 5 interns.

Molecular Biology NSF Fellow, *University of Georgia, Dr. Travis Glenn* *05-09/2005*

- Conducted independent research on pair bonding in alligators and determining mutation rates by carrying out ABI Prism sequencing for SNP and microsatellites, multiplexed PCR, and sequence analyzes for genotyping.

Genetics Research Assistant, *Evergreen State College, Dr. Erik Thusen* *2004–2005*

- Independent research on the population genetics of moon jellies, gaining experience in genomic DNA purification, PCR for species identification, and phylogenetic analyzes.

Developmental Biology Research Assistant, *Evergreen State College, Dr. Donald Morisato* *2003–2004*

- Investigated the molecular mechanisms of generating the anterior-posterior axis in *Drosophila,* by carrying out *lac z* staining, *in situ* hybridization, and genetic crosses.

Environmental Science Research Assistant, *Pacific Crest Community School, Dr. Tigerin Peare* *2000–2001*

- Independent research on the *E. coli* levels in the Willamette River, Portland, OR. Data collected results in new city testing and regulations on the city water treatment facilities.

Teaching/Mentoring Experience

Research Mentor for Undergraduate Students, *UC Santa Barbara* *2008-2011*

- Lead undergraduate students in research projects; taught techniques and guided experiments.

Teaching Assistant for epigenetics, developmental & biology laboratory, *UCSB, intermittently 2007-2010*

- Prepared lectures, gave lectures, and instructed laboratory sections.

Teaching Assistant, *Santa Barbara City College,* *2006 - 2007*

- Molecular biology, botany and general biology laboratories.

Scientific Computing Specialist, *Evergreen State College* *2003 –2006*

- Taught and helped students with science software - including biological modeling, statistics, phylogenetic, mathematical, and graphical programs.

Publications

Holz-Schietinger, C., Matje, D.M., Reich, N.O. Mutations in DNA methyltransferase (DNMT3A) observed in Acute Myeloid Leukemia patients disrupt methylation of clustered CpG sites. *J. Biol. Chem, 287: 30941*-30951 (2012).

Holz-Schietinger, C., and Reich, N.O. RNA modulation of the human *de novo* methyltransferase 3A (DNMT3A). *Nucleic Acids Res.* 40: 8550-8557 (2012).

Holz-Schietinger, C., Matje, D.M., Flexer Harrison, M. and Reich, N.O. DNA oligomerization controls the mechanism of *de novo* DNA methylation. *J. Biol. Chem.* 286, 41479-41488 (2011).

Holz-Schietinger, C. & Reich, N.O. The inherent processivity of the human *de novo* methyltransferase 3A (DNMT3A) is enhanced by DNMT3L. *J. Biol. Chem.* 285, 29091–29100 (2010).

Holz-Schietinger, C., Matje, D.M. and Reich, N.O. *De novo* DNA methyltransferase DNMT3A: regulation of oligomeric state and mechanism of action in response to physiological pH changes. *Biochimica et Biophysica Acta (BBA). 1850:1131-1139 (2015).*

Purdy, M.M., **Holz-Schietinger, C**. & Reich, N.O. Identification of a second DNA binding site in human DNA methyltransferase 3A by substrate inhibition and domain deletion. *Arch. Biochem. Biophys.* 498, 13-22 (2010).

Lance, S., Tuberville, T., Dueck, L., **Holz-Schietinger, C.,** Trosclair, P., Elsey, R., Glenn, T. Multi-year multiple paternity and mate fidelity in the American alligator, *Alligator mississippiensis. Molecular Ecology.* 2009. 18, 4508-4520 (2009).

Awards/Fellowships

- 2010-2011 Philip & Aida Siff Fellowship for excellent undergraduate student mentoring
- 2010, 2011 Best graduate presentation for USCB BMSE and MCDB departments
- 2009 Honorable Mention NSF Graduate Research Fellowship
- 2007-2008 Amgen Fellowship
- 2005 NFS Undergraduate Research Fellowship
- 2005 Evergreen foundation grant (research funding)
- 2002-2004 Evergreen State College foundation scholarship for academic excellence
- 2001-2002 USAA All-American Scholar, Philip S. Wilder Award for Academic Distinction, National Deans List

Training Courses /Research Conferences

- 14th Weurman Flavour Research Symposium, (2014) Cambridge, UK
- FKS Flavor Reaction Course, (2014), University of Missouri St Louis
- American Oil Chemistry Society (AOCS) conference, Lecithin Functions in Technology and Nutrition Course, (2013) AOCS Montreal QC, Canada
- Enzymes, Coenzymes & Metabolic Pathways Gordon conference, (2011, 2010) Waterville Valley, NH

Sample	Concentration (ppb) of Compounds in Headspace of Cooked Beef											Odor Activity Value (OAV)				Odor Descriptor
	Control	Variant 1	Variant 2	Variant 3	Variant 4	Variant 5	Variant 6	Variant 7	Variant 8	Variant 9	Variant 10	Control	Variant 1	Variant 2	Variant 3	
Lactones																
2(3H)-Furanone, 5-ethyldihydro-	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	24,918	ND	ND	ND	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
3(2H)-Furanone, dihydro-2-methyl-	ND	ND	ND	ND	70,307	134,098	90,684	78,528	41,167	70,039	52,986	1.9	1.1	1.0	0.8	www.thegoodscentscompany.com
Butyrolactone	15,855	2,514	ND	9,093	43,283	414,965	344,878	325,440	254,578	55,507	172,275	9.2	7.5	1.3	3.8	Resconi et al. (2013, Molecules, 18:6748-81)
Furfural	ND	ND	ND	16,182	831,028	2,557,660	2,842,847	5,321,805	21,910,846	33,076,245	26,952,262	3.1	6.4	39.8	32.4	Resconi et al. (2013, Molecules, 18:6748-81)
2(3H)-Furanone, dihydro-5-pentyl-	ND	ND	ND	ND	ND	53,367	ND	ND	ND	ND	ND	Novel	ND	ND	ND	Song et al. (2012, J. Chromatog. A, 1254:115-24); Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
O- and N-Heterocyclic compounds																
Furan, 2-pentyl-	169,688	756,003	1,074,667	3,234,344	354,931	710,485	2,330,673	4,908,319	5,546,200	5,863,327	6,376,371	1.5	11.7	13.5	8.9	Song et al. (2012, J. Chromatog. A, 1254:115-24); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com; Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8)
Furan, 2-methyl-	ND	ND	ND	1,305,496	6,920,678	15,759,524	6,744,937	13,398,632	17,639,903	19,782,037	23,741,932	2.3	1.9	2.9	3.2	Song et al. (2012, J. Chromatog. A, 1254:115-24); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com
2-pentyl-Pyridine	ND	ND	ND	ND	ND	ND	ND	ND	143,621	90,947	77,675	ND	ND	Novel	Novel	Mottram (1998, Food Chem., 62:415-24)
2-ethyl-Furan	ND	261,266	446,522	1,298,861	123,564	1,532,213	840,812	1,872,296	1,283,533	1,230,129	1,404,690	12.4	13.0	6.3	0.9	www.thegoodscentscompany.com
Pyrazine, 2-ethyl-5-methyl-	ND	ND	25,319	137,355	ND	ND	ND	ND	181,741	226,579	457,241	ND	ND	Novel	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Pyrazine, 2,5-dimethyl-	ND	ND	ND	87,197	ND	ND	ND	301,563	2,615,368	3,346,920	5,407,658	ND	Novel	Novel	Novel	www.thegoodscentscompany.com; Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Pyrazine, methyl-	ND	458,644	808,465	5,104,659	248,740	370,185	721,844	3,671,282	27,253,089	30,915,735	42,337,597	1.5	12.9	121.0	149.7	Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Indole	ND	ND	ND	40,013	ND	ND	ND	39,279	109,587	133,293	150,508	ND	Novel	Novel	Novel	www.thegoodscentscompany.com
S-Heterocyclic compounds																
2-Acetylthiazole	54,332	ND	ND	ND	1,689,545	931,426	1,149,078	14,455,827	173,370,592	361,866,807	460,608,605	0.5	8.6	214.2	272.6	Song et al. (2012, J. Chromatog. A, 1254:115-24); McGorrin et al. (2001, Special Publication Royal Soc. Chem., 274:67-84); www.thegoodscentscompany.com; Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8)
2-Furfurylthiol	ND	ND	ND	ND	298,298	886,091	895,895	476,371	ND	ND	ND	3.0	1.6	ND	ND	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Farmer & Patterson (1991, Food Chem., 40:201-5)
2-Methyl-3-furanthiol	ND	ND	ND	ND	323,141	39,001,950	60,841,403	103,481,535	98,624	212,762	301,787	121	320	0.7	0.0	Song et al. (2012, J. Chromatog. A, 1254:115-24); www.thegoodscentscompany.com; Mottram (1998, Food Chem., 62:415-24)
3-Thiophenemethanol	ND	ND	ND	ND	ND	ND	ND	ND	197,893	144,691	191,824	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Farmer & Patterson (1991, Food Chem., 40:201-5)
3-Methyl-2-thiophenecarboxaldehyde	ND	ND	ND	ND	314,590	623,650	659,737	952,378	1,116,277	977,661	1,402,630	2.0	3.0	3.1	4.5	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
Ethanone, 1-(4,5-dihydro-2-thiazolyl)-	ND	ND	ND	ND	ND	ND	ND	83,360	616,786	503,970	761,123	ND	ND	Novel	Novel	www.thegoodscentscompany.com;
5-Methyl-2-thiophenecarboxaldehyde	125,170	75,629	102,226	338,545	70,308	191,517	105,220	611,651	6,520,866	25,169,753	103,060,745	0.9	7.6	356.5	1,461.0	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
3-methyl-Thiophene	ND	ND	ND	ND	ND	210,710	283,961	123,128	1,627,609	1,854,428	4,319,324	Novel	Novel	Novel	Novel	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
Thiophene	ND	ND	ND	6,914	3,532,628	2,892,410	6,025,778	27,079,671	60,644,016	37,690,815	40,098,096	0.8	7.7	10.7	11.3	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
3(2H)-Thiophenone, dihydro-2-methyl-	ND	ND	ND	ND	138,043	104,767	129,130	852,867	2,028,273	1,231,819	2,248,689	0.8	6.2	8.9	16.3	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
Dihydro-3-(2H)-thiophenone	ND	ND	ND	ND	23,556	20,083	32,274	110,406	523,436	364,729	708,356	0.9	4.7	15.5	30.1	Song et al. (2012, J. Chromatog. A, 1254:115-24); McGorrin et al. (2001, Special Publication Royal Soc. Chem., 274:67-84); www.thegoodscentscompany.com; Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8)
Benzothiazole	ND	1,905	ND	19,507	9,177	23,501	15,082	19,921	33,597	74,262	75,346	2.6	2.0	8.1	6.1	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-ethyl-Thiophene	ND	ND	ND	ND	ND	54,572	43,576	115,064	176,302	145,384	243,368	Novel	Novel	Novel	Novel	www.thegoodscentscompany.com
2-methyl-5-propyl-Thiophene	ND	ND	ND	ND	ND	17,454	14,109	23,104	20,718	23,869	28,288	Novel	Novel	Novel	Novel	
4-(Methylthio)thiophenol	ND	ND	ND	ND	ND	10,324	7,534	68,670	103,332	21,344	12,513	Novel	Novel	Novel	Novel	
3-Thiophenecarboxaldehyde	138,681	44,299	29,330	76,721	25,501	383,473	194,595	6,852,026	92,565,186	91,829,374	110,121,055	9.6	267.0	3,599.9	4,315.3	
5-Thiazoleethanol, 4-methyl-	ND	ND	ND	ND	534,899	825,826	566,918	1,482,521	776,664	851,847	2,046,842	1.5	2.8	1.6	3.8	www.thegoodscentscompany.com
2,4-dimethyl-Thiazole	ND	ND	ND	ND	ND	ND	ND	ND	1,190,614	663,344	675,389	ND	ND	Novel	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2,4,5-trimethyl-Thiazole	ND	ND	ND	ND	34,061	81,739	98,355	162,291	254,522	179,635	154,982	2.4	4.8	5.3	4.6	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Carboxylic Acids																
Hexanoic acid	ND	ND	ND	ND	ND	70,367	58,992	60,166	58,027	54,875	56,525	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Sell et al. (2009, Food Chem., 114:317-22)
Octanoic acid	35,160	ND	ND	ND	ND	71,688	ND	ND	ND	50,585	ND	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308)
Hexadecanoic acid	21,212	378,491	572,236	1,346,727	ND	ND	42,866	91,670	221,214	507,170	1,512,436	ND	Novel	Novel	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
Propanoic acid	ND	ND	ND	ND	ND	64,956	59,385	63,182	25,718	33,235	53,568	Novel	Novel	Novel	Novel	www.thegoodscentscompany.com
Aldehydes																
Decanal	57,844	870,579	1,382,206	2,755,248	ND	33,767	37,263	115,149	470,376	1,095,986	2,908,319	Novel	Novel	Novel	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97); Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
Undecanal	ND	0	0	2,525	ND	1,090	356	45,163	180,604	220,641	224,669	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
Butanal	ND	ND	ND	118,902	ND	2,388,522	2,830,675	2,256,136	3,648,224	5,653,384	5,678,285	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Heptanal	90,245	138,393	224,287	406,921	ND	78,344	127,417	91,988	156,024	269,810	498,125	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Propanal, 2-methyl-	125,022	1,386,906	2,068,389	5,835,755	ND	80,686	73,087	501,617	16,911,440	31,685,291	39,771,862	Novel	Novel	Novel	Novel	Guth & Grosch (1995, Z. Lebensm Unters Forsch, 201:25-9)
Benzaldehyde, 4-ethyl-	ND	ND	1,655	ND	8,583	33,024	26,367	22,926	21,553	13,037	15,069	3.8	2.7	1.3	1.8	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)

Propanal	NA	ND	49,776	448,102	ND	270,076	211,974	291,138	2,371,841	3,349,197	3,435,023	Novel	Novel	Novel	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Hexanal	75,075	95,183	112,957	325,332	ND	ND	20,228	32,489	227,058	191,953	302,062	ND	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Rochat et al. (2007, J. Chromatogr., 1147:85-94)
Butanal, 3-methyl-	557,072	18,263,998	31,231,931	110,254,766	ND	2,362,618	2,533,443	12,904,160	202,805,346	308,133,887	327,395,687	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Rochat & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85)
Pentanal	ND	112,415	ND	719,891	ND	ND	ND	ND	ND	1,199,001	1,334,055	ND	ND	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308)
3-methyl-2-Butenal	ND	92,647	147,360	220,813	ND	153,253	60,188	1,457,962	27,806,744	26,383,307	18,675,745	Novel	Novel	Novel	Novel	www.thegoodscentscompany.com
Sulfides																
Dimethyl sulfide	NA	ND	16,805	138,735	ND	608,212	4,277	46,842	175,231	115,614	363,092	Novel	Novel	Novel	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Grigorakis et al. (2003, Aquaculture, 225:109-19); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
Dimethyl trisulfide	9,651	625,067	696,993	1,561,353	ND	ND	ND	8,894	49,681	145,651	427,713	ND	Novel	Novel	Novel	Rochat et al. (2007, J. Chromatogr., 1147:85-94); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Disulfide, dimethyl	166,715	2,216,160	3,343,933	6,841,436	ND	ND	ND	17,319	125,598	163,733	1,107,514	ND	Novel	Novel	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Methanethiol	ND	ND	ND	ND	ND	13,850	68,075	1,032,552	15,252	ND	ND	Novel	Novel	ND	ND	Moon et al. (2006, Food Res. Int., 39:294-308); Guth & Grosch (1995, Z. Lebensm Unters Forsch, 201:25-6); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Ketones																
2-Butanone	422,992	10,234,816	15,836,368	58,951,787	1,801,674	4,793,578	5,923,348	39,080,140	174,676,835	246,577,952	360,484,573	2.4	16.0	128.1	167.4	Moon et al. (2006, Food Res. Int., 39:294-308); Rochat & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85)
2-Nonanone	62,751	403,604	810,729	3,070,051	ND	63,605	76,414	94,606	204,317	599,275	1,663,501	Novel	Novel	Novel	Novel	Rochat & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-Undecanone	43,609	963,841	1,575,957	3,492,050	3,719	82,402	87,638	204,052	703,973	1,491,956	3,177,597	10.4	Decrease	Decrease	Decrease	Rochat & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2,3-Butanedione	566,203	523,951	45,103,463	46,719,319	950,004	2,282,634	2,136,841	8,438,865	246,427,606	658,788,553	755,251,721	1.8	8.3	646.0	619.0	Rochat & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-Heptanone	69,010	129,443	183,580	549,173	ND	59,309	160,170	154,193	129,189	115,276	262,944	Novel	Novel	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Rochat & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Acetophenone	41,840	111,440	169,138	564,399	28,223	105,993	88,379	189,534	570,870	399,314	708,004	2.3	2.8	8.2	5.1	www.thegoodscentscompany.com
2,3-Pentanedione	ND	333,214	627,685	2,199,141	ND	39,380	14,589	98,573	700,537	999,962	2,173,388	Novel	Novel	Novel	Novel	www.thegoodscentscompany.com
Alcohols																
Furaneol	ND	295,105	385,068	1,925,164	ND	ND	ND	ND	47,673	153,459	774,672	ND	ND	Novel	Novel	Guth & Grosch (1995, Z. Lebensm Unters Forsch, 201:25-6); Moon et al. (2006, Food Res. Int., 39:294-308); Rochat et al. (2007, J. Chromatogr., 1147:85-94); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Farmer
2-Ethyl-1-hexanol	ND	ND	ND	ND	311,874	1,016,073	820,983	ND	755,739	ND	424,100	3.3	ND	ND	1.4	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)

DECLARATION UNDER 37 CFR §1.132

I, Celeste Holz-Schietinger, Ph.D., hereby declare as follows:

1. I am currently a Principal Scientist at Impossible Foods in Redwood City, California. I have held this position since 2015, and I am responsible for leading the Flavor Department at Impossible Foods. In this role, I direct flavor technology research as well as flavor product integration. I began working for Impossible Foods in 2012 as a Scientist, and I was elevated to a Senior Scientist in 2013. While at Impossible Foods, I have been the lead chemist on the identification, characterization and manipulation of biological flavor catalysis and potentiation, including through the use of heme-containing proteins.

2. I received my Bachelors of Science (B.S.) degree in Molecular Biology and Evolution, with a minor in Chemistry, in 2005 from The Evergreen State College in Olympia, Washington, and I received my Doctor of Philosophy (Ph.D.) degree in Biochemistry from the University of California at Santa Barbara in 2012. I was an Enzymology Research Associate at the University of California at Santa Barbara from 2007 to 2012, and I also was a Biochemistry Visiting Associate at Stanford University in 2009.

3. A copy of my curriculum vitae is attached as **Exhibit A**.

4. Gas Chromatography-Mass Spectroscopy (GCMS) and Gas Chromatography-Olfactory (GCO) methods were used to evaluate the production of volatile compounds during cooking of flavor reaction mixtures, and in particular, to evaluate the production of volatile compounds that are associated with meaty aroma. The first flavor reaction mixture contained 0.5% (note 0.5% = 5 mg/mL) leghemoglobin (LegH); the second contained 10 mM ribose with 1.5 mM thiamine (ribose + thiamine 1.5 mM); the third contained 0.5% leghemoglobin with 10 mM ribose with 1.5 mM thiamine (LegH + ribose + thiamine 1.5 mM); the fourth contained 10 mM ribose with 1.5 mM cysteine (ribose + cysteine 1.5 mM); the fifth contained 0.5% leghemoglobin with 10 mM ribose with 1.5 mM cysteine (LegH + ribose + cysteine 1.5 mM). All flavor reaction mixtures were adjusted to pH 6.5 using 50 mM of potassium phosphate buffer and 100 mM of sodium chloride buffer. Two mL of each flavor reaction mixture were prepared in 10 mL GCMS glass vials (referred to as "flavor reaction mixture samples" or "samples" hereinafter). Each sample was analyzed in duplicate.

5. An Agilent 7890A GC system coupled with a Leco TruTOF HT system was used along with a Gerstel MultiPurpose Sampler for auto-sampling. The Gerstel auto-sampler was used to cook, cool, and perform HS-SPME on each of the samples. Each sample was heated to 150°C for 5 minutes with 750 rpm of agitation to "cook" the material. This was followed by a two-phase active cooling step that decreased the agitator temperature from 150°C to 50°C. The sample was extracted for 20 minutes at 50°C with 500 rpm of agitation using a 50/30 µm Divinylbenzene / Carboxen / Polydimethylsiloxane (DVB/CAR/PDMS), Stable flex, 23 Ga, Autosampler (Supleco, Cat# 57298-U) fiber.

6. Extracted samples were run on the GCMS by desorbing the SPME fiber in the Gerstel controlled CIS inlet for 6 minutes at 240°C. The desorbed sample was separated on a 60-meter wax column (Agilent, VF-WAXms 60 m x 0.25 mm x 0.25 mm, Part# CP9207) using a 35 minutes GCMS method (35°C for 2 minutes, ramp of 5C°/min until 172°C, ramp of 60C°/min until 255°C, 255°C for 4 minutes) and 1.75 mL/min Helium flow rate and splitless mode. Separated compounds were analyzed by the mass spectrometer, and all data was collected over a 14-500 µ mass range, with an acquisition rate of 10 spectra/sec, and a detector voltage off-set of 100.

7. Samples were then analyzed using Leco ChromaTOF optimized for TruTOF HT (Version 4.51.6.0) coupled with the NIST MS Search 2.2. The identity of each peak in each sample was identified in a two-step process. First, the mass spectrum of each peak with a signal to noise greater than 50 was matched to a mass spectrum in the NIST library using a match threshold of 650. Additionally, an internally developed calibration method was applied to the data set to confirm the identity of the compounds of interest. In the second step, the Statistical Compare function of ChromaTOF was used to align the named analytes across all samples in a set. The criteria for aligning a single peak across all samples were a similarity score (similarity between the peak spectra across all samples) of 700, a maximum retention time difference of 3 seconds, and the peak must have been present in at least two samples.

8. The amount of each aroma compound was determined by averaging the signal intensities from the duplicate experiments. The amount of each aroma compound that can be

attributed to leghemoglobin (LegH)[1] was determined by using the average signal intensity for each aroma compound produced in the LegH sample as a baseline to standardize the average signal intensity for the same aroma compound produced in the LegH + ribose + thiamine 1.5 mM sample and/or the LegH + ribose + cysteine 1.5 mM sample. The fold-change in each aroma compound was determined by dividing the signal intensity produced in the presence of LegH (i.e., in the LegH + ribose + thiamine 1.5 mM sample) by the signal intensity produced in the absence of LegH (i.e., in the ribose + thiamine 1.5 mM sample); or the fold-change in each aroma compound was determined by dividing the signal intensity produced in the presence of LegH (i.e., in the LegH + ribose + cysteine 1.5 mM sample) by the signal intensity produced in the absence of LegH (i.e., in the ribose + cysteine 1.5 mM sample). If the signal intensity in the absence of LegH (i.e., in the ribose + thiamine 1.5 mM sample *or* in the ribose + cysteine 1.5 mM sample) was not detectable, the aroma compound produced in the presence of LegH (i.e., in the LegH + ribose + thiamine 1.5 mM sample *or* in the LegH + ribose + cysteine 1.5 mM sample) was indicated as "novel," meaning that the aroma compound was newly produced (to the limits of detection of the technique) in the LegH + ribose +thiamine (or cysteine) sample.

9. The results of the above-described experiments are shown in **Exhibit B**. The first column shows the list of aroma compounds, grouped into six (6) different classes of chemicals. The next columns show the amount of each aroma compound, calculated as described above, for each of the samples (LegH; ribose + thiamine 1.5 mM; LegH + ribose + thiamine 1.5 mM; ribose + cysteine 1.5 mM; and LegH + ribose + cysteine 1.5 mM) followed by columns indicating the fold-change, calculated as described above, or indicating that the aroma compound is novel as described above. The next column provides, if available, one or more peer-reviewed publications in which that particular aroma compound is associated with meaty aroma.

10. The presence of LegH in combination with thiamine resulted in the production of **twenty five (25)** novel meat associated aroma compounds that were not produced in the absence of LegH. The 25 different novel aroma compounds can be divided into six (6) different classes of compounds as follows.

- *Ketones*: three (3) novel ketones were produced (i.e., 2-butanone, 2-heptanone, 3-octanone);

[1] Aroma compounds that can be attributed to LegH refer to compounds that are, independently, either not detectably produced or produced in a detectably smaller amount upon cooking compared with the control samples (e.g., LegH-alone or Ribose + Cysteine in the absence of LegH).

- *Sulfur (S)-containing heterocyclic*: four (4) novel S-containing heterocyclic compounds were produced (i.e., 2-methyl-3-furanthiol, 2-methyl-thiazole, thiazole, 4-methyl-5thiazoleethanol);
- *Sulfides*: no (0) novel sulfide compounds were produced;
- *Aldehydes*: nine (9) novel aldehydes were produced (i.e., 2-methyl-propanal, pentanal, nonanal, undecanal, 3-methyl-butanal, furfural, decanal, 3-methyl-2-butenal, octanal);
- *Alcohols*: two (2) novel alcohol was produced (i.e., 1-hexanol, 2-furanmethanol); and
- *Oxygen (O)- and/or Nitrogen (N)-containing heterocyclic*: seven (7) novel O- and/or N-containing heterocyclic compounds were produced (i.e., 2-acetyl-1-pyrroline, methyl-pyrazine, 2-methyl-furan, pyrazine, 2-pentyl-furan, 2,5-dimethyl-pyrazine, 2,3-dimethyl-pyrazine).

Significantly, each of the twenty five (25) different novel aroma compounds have been associated, directly or indirectly, with meaty aroma; see, for example, the references provided in **Exhibit B**. For example, 2-methyl-3-furanthiol[2,3], 2-methyl-thiazole, thiazole, 4-methyl-5-

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[2] See also the enclosed Declaration under 37 CFR § 1.132 by Dr. Keith R. Cadwallader ("the Cadwallader Declaration"), attached as **Exhibit C**, paragraph 15, which states:

> Among the 28 compounds identified in the [Holz-Schietinger Declaration dated December 2, 2016 as filed in U.S. Application Serial No. 14/797,006] experiments, two in particular are well-known to possess meaty sensory characteristics and to be important and potent odorants in cooked meat. Specifically, 2-methyl-3-furanthiol (MFT) and its dimerized form bis(2-methyl-3-furyl)disulfide (MFT-MFT) are important aroma compounds in cooked meat (Tang et al., 2013). Both compounds have extremely low odor detection thresholds of 0.7 parts-per-trillion (pptr) and 0.02 pptr, respectively (Belitz et al. 2006), and are usually only present in low abundance (pptr range) in cooked meat (Tang et al., 2013). Despite their low abundance in meat they are still among the most impactful aroma compounds because of their extremely low odor detection thresholds.

[3] See also the enclosed Declaration under 37 CFR § 1.132 by Dr. Devin Grant Peterson ("the Peterson Declaration"), attached as **Exhibit D**, paragraph 21, which states:

> Additionally, the [Holz-Schietinger Declaration dated December 2, 2016 as filed in U.S. Application Serial No. 14/797,006] demonstrates that 2-methyl-3-furanthiol (MFT) and the major degradation product of MFT, the dimer, bis(2-methyl-3-furyl) disulfide, were detected only in the aqueous thermal reaction samples that contained Leghemoglobin. MFT is a very potent meaty odorant, with an extremely low odor threshold at 0.007 µg/kg in water, and is known to have an important contribution to meat aroma. See, for example, Kerscher and Grosch (1998, J. Agric. Food Chem., 46:1954-58); Tang et al. (2013, J. Sulfur Chem., 34:38-47); and Withcombe and Mussinan (1988, J. Food Sci., 53:658-60). It is surprising to me that an intense meaty aroma compound such as MFT was uniquely identified in the reaction mixture upon the addition of LegH.

thiazoleethanol nonanal, undecanal, furfural, decanal, 2-acetyl-1-pyrroline, methyl-pyrazine, and 2,3-dimethyl-pyrazine have been identified as being very important for meaty aroma.

11. The presence of LegH in the flavor reaction mixture with 10 mM ribose and 1.5 mM thiamine also resulted synergistic and dramatic increase in the amount at least **twelve (12)** meat associated aroma compounds produced upon heating compared to the amount of the same aroma compounds produced upon heating in the absence of LegH (i.e., in the ribose + thiamine 1.5 mM sample) or in the LegH only sample. For example, in the presence of LegH, the amount of:

- 2,3-butanedione was increased **36-fold**;
- 2,3-pentanedione was increased **13-fold**;
- dimethyl trisulfide was increased **40-fold**;
- carbon disulfide was increased **19-fold**;
- acetaldehyde was increased **36-fold**;
- 2-methyl-butanal was increased **312-fold**; and
- benzaldehyde was increased **161-fold**.

Significantly, 2,3-butanedione and 2,3-pentanedione have been strongly associated with meaty aroma. See, for example, the references cited in **Exhibit B**.

12. The presence of LegH in combination with cysteine resulted in the production of **twenty two (22)** novel aroma compounds that were not produced in the absence of LegH. The 22 different novel meat associated aroma compounds can be divided into six (6) different classes of compounds as follows.

- *Ketones*: three (3) novel ketones were produced (i.e., 2-heptanone, 3-octanone, 2,3-pentanedione);
- *Sulfur (S)-containing heterocyclic*: five (5) novel S-containing heterocyclic compounds were produced (i.e., 2-methyl-3-furanthiol, 2-methyl-thiazole, dihydro-3-(2H)-thiophenone, 3-thiophenemethanol, 3-methyl-thiophene);
- *Sulfides*: no (0) novel sulfide compounds were produced;
- *Aldehydes*: seven (7) novel aldehydes were produced (i.e., pentanal, butanal, nonanal, furfural, decanal, 3-methyl-2-butenal, octanal);
- *Alcohols*: one (1) novel alcohol was produced (i.e., 1-hexanol).
- *Oxygen (O)- and/or Nitrogen (N)-containing heterocyclic*: six (6) novel O- and/or N-containing heterocyclic compounds were produced (i.e., 2-acetyl-1-pyrroline,

methyl-pyrazine, 2-methyl-furan, 2-pentyl-furan, 2,5-dimethyl-pyrazine, 2,3-dimethyl-pyrazine).

Significantly, each of the twenty two (22) different novel aroma compounds have been associated, directly or indirectly, with meaty aroma; see, for example, the references provided in Exhibit B. For example, 2,3-pentanedione, 2-methyl-3-furanthiol, 2-methyl-thiazole, dihydro-3-(2H)-thiophenone, 3-thiophenemethanol, nonanal, furfural, decanal, 2-acetyl-1-pyrroline, methyl-pyrazine, and 2,3-dimethyl-pyrazine have been identified as being very important for meaty aroma.

13. The presence of LegH in the flavor reaction mixture with 10 mM ribose and 1.5 mM cysteine also resulted synergistic and dramatic increase in the amount at least **seventeen (17)** meat associated aroma compounds produced upon heating compared to the amount of the same aroma compounds produced upon heating in the absence of LegH (i.e., in the ribose + cysteine 1.5 mM sample) or in the LegH only sample. For example, in the presence of LegH, the amount of:

- 2,3-butanedione was increased **33-fold**;
- 2-butanone was increased **23-fold**;
- thiazole was increased **47-fold**;
- 2-acetylthiazole was increased **35-fold**;
- thiophene was increased **30-fold**;
- carbon disulfide was increased **23-fold**;
- 2-methyl-propanal was increased **46-fold**;
- benzaldehyde was increased **29-fold**;
- undecanal was increased **11-fold**;
- 3-methyl-butanal was increased **56-fold**;
- 2-methyl-2-butenal was increase **31-fold**; and
- pyrazine was increased **21-fold**.

Significantly, 2,3-butanedione, 3-methyl-butanal, 2-acetylthiazole and thiophene have been strongly associated with meaty aroma. See, for example, the references cited in **Exhibit B**.

15. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are

punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

Respectfully submitted,

Date: 8/17/17

Celeste Holz-Schietinger

EXHIBIT A

Celeste Holz-Schietinger, PhD

Principal Scientist, Impossible Foods

Redwood City, CA
celeste.holz@impossiblefoods.com

Innovative R&D Scientist and Product Developer responsible for leading the Flavor Department at Impossible Foods. Technical skills include enzymology, flavor chemistry, sensory science, lipid and protein biochemistry, protein purification, analytical chemistry, metabolic pathways and small molecule identification.

Education

Ph.D. Biochemistry, **University of California** at Santa Barbara — *2007-2012*
from the Biomolecular Science and Engineering Program

B.S. Molecular Biology & Evolution, minor in Chemistry, — *2001- 2005*
The Evergreen State College, Olympia, WA

Research

Principal Scientist, *Impossible Foods, Dr. Pat Brown* — *2015- Current*

Senior Scientist, *Impossible Foods, Dr. Peg Scott, Dr. Chris Davis* — *2013- 2015*

Scientist, *Impossible Foods, Dr. Chris Davis* — *2012-2013*

- R&D scientist and flavor department leader at Impossible Foods, directing flavor technology research and flavor product integration.
- Manage a team of scientists to understand the fundamental biology and chemistry of flavor perception and flavor creation. We identify the catalysis and precursors that create aroma compounds, determine how aroma compounds create particular flavors profiles, and then how to recreate these flavors in products.
- Lead chemist on the identification, characterization and manipulation of biological flavor catalysis, including heme-proteins, microbes, and enzymes.
- Support off flavor identification and removal in products and feedstock with product developers, protein biochemist and process engineers.
- Flavor product integration, leading efforts for creating meat replicas from plants that taste and smell like animal based meats, working closely with manufacturing to scale the process, and marketing to meet consumers desires.
- Previous roles also included but not limited to sensory panel leader, lipid group leader, and product developer creating nut-base cheese (Kite Hill Cheese).

Enzymology Research Associate, *UC Santa Barbara, Dr. Norbert Reich* — *2007 –2012*

- Researched the underlying chemical mechanisms that regulate how the genetic code is translated to creates phenotypic variation in human cells; particularly, understanding the developmental enzymes (DNMT3B and DNMT3A) and their modulators.
- Identified the mechanisms and structural changes that occurred with mutated forms of DNMT3A, highly associated with diseased states that change gene expression.

- Particular specialty was the development and execution of kinetic and biophysical assays to characterize the enzymology and regulation of the *de novo* DNA methyltransferase (DNMT3A).
- Mentored and lead 6 undergraduate researchers.

Biochemistry Visiting Associate, *Stanford University, Dr. Pat Brown (collaboration 2008-2010) 06-11/2009*

- Global identification of RNA associated with developmental enzymes (DNMT3B and DNMT3A) through cellular differentiation and correlated to gene expression and DNA methylation patterns. Developed RNA library construction protocols for next generation sequencing, and became skilled in microarray, hES cell culture, and fluorescence microscopy.

Scientist in Endocrinology, *U.S. Geological Survey* *05-09/2006*

- Conducted research on the stress response of translocation on the desert tortoise, though ELISA-based assays, leading team of 5 interns.

Molecular Biology NSF Fellow, *University of Georgia, Dr. Travis Glenn* *05-09/2005*

- Conducted independent research on pair bonding in alligators and determining mutation rates by carrying out ABI Prism sequencing for SNP and microsatellites, multiplexed PCR, and sequence analyzes for genotyping.

Genetics Research Assistant, *Evergreen State College, Dr. Erik Thusen* *2004–2005*

- Independent research on the population genetics of moon jellies, gaining experience in genomic DNA purification, PCR for species identification, and phylogenetic analyzes.

Developmental Biology Research Assistant, *Evergreen State College, Dr. Donald Morisato* *2003–2004*

- Investigated the molecular mechanisms of generating the anterior-posterior axis in *Drosophila,* by carrying out *lac z* staining, *in situ* hybridization, and genetic crosses.

Environmental Science Research Assistant, *Pacific Crest Community School, Dr. Tigerin Peare* *2000–2001*

- Independent research on the *E. coli* levels in the Willamette River, Portland, OR. Data collected results in new city testing and regulations on the city water treatment facilities.

Teaching/Mentoring Experience

Research Mentor for Undergraduate Students, *UC Santa Barbara* *2008-2011*

- Lead undergraduate students in research projects; taught techniques and guided experiments.

Teaching Assistant for epigenetics, developmental & biology laboratory, *UCSB, intermittently 2007-2010*

- Prepared lectures, gave lectures, and instructed laboratory sections.

Teaching Assistant, *Santa Barbara City College,* *2006 - 2007*

- Molecular biology, botany and general biology laboratories.

Scientific Computing Specialist, *Evergreen State College* *2003 –2006*

- Taught and helped students with science software - including biological modeling, statistics, phylogenetic, mathematical, and graphical programs.

Publications

Holz-Schietinger, C., Matje, D.M., Reich, N.O. Mutations in DNA methyltransferase (DNMT3A) observed in Acute Myeloid Leukemia patients disrupt methylation of clustered CpG sites. *J. Biol. Chem, 287: 30941*-30951 (2012).

Holz-Schietinger, C., and Reich, N.O. RNA modulation of the human *de novo* methyltransferase 3A (DNMT3A). *Nucleic Acids Res.* 40: 8550-8557 (2012).

Holz-Schietinger, C., Matje, D.M., Flexer Harrison, M. and Reich, N.O. DNA oligomerization controls the mechanism of *de novo* DNA methylation. *J. Biol. Chem.* 286, 41479-41488 (2011).

Holz-Schietinger, C. & Reich, N.O. The inherent processivity of the human *de novo* methyltransferase 3A (DNMT3A) is enhanced by DNMT3L. *J. Biol. Chem.* 285, 29091–29100 (2010).

Holz-Schietinger, C., Matje, D.M. and Reich, N.O. *De novo* DNA methyltransferase DNMT3A: regulation of oligomeric state and mechanism of action in response to physiological pH changes. *Biochimica et Biophysica Acta (BBA). 1850:1131-1139 (2015).*

Purdy, M.M., **Holz-Schietinger, C**. & Reich, N.O. Identification of a second DNA binding site in human DNA methyltransferase 3A by substrate inhibition and domain deletion. *Arch. Biochem. Biophys.* 498, 13-22 (2010).

Lance, S., Tuberville, T., Dueck, L., **Holz-Schietinger, C.,** Trosclair, P., Elsey, R., Glenn, T. Multi-year multiple paternity and mate fidelity in the American alligator, *Alligator mississippiensis. Molecular Ecology.* 2009. 18, 4508-4520 (2009).

Awards/Fellowships

- 2010-2011 Philip & Aida Siff Fellowship for excellent undergraduate student mentoring
- 2010, 2011 Best graduate presentation for USCB BMSE and MCDB departments
- 2009 Honorable Mention NSF Graduate Research Fellowship
- 2007-2008 Amgen Fellowship
- 2005 NFS Undergraduate Research Fellowship
- 2005 Evergreen foundation grant (research funding)
- 2002-2004 Evergreen State College foundation scholarship for academic excellence
- 2001-2002 USAA All-American Scholar, Philip S. Wilder Award for Academic Distinction, National Deans List

Training Courses /Research Conferences

- 14th Weurman Flavour Research Symposium, (2014) Cambridge, UK
- FKS Flavor Reaction Course, (2014), University of Missouri St Louis
- American Oil Chemistry Society (AOCS) conference, Lecithin Functions in Technology and Nutrition Course, (2013) AOCS Montreal QC, Canada
- Enzymes, Coenzymes & Metabolic Pathways Gordon conference, (2011, 2010) Waterville Valley, NH

Compound	Signal Intensity: ND = Not detected (signal to noise of 50, signal of > 10,000)					Fold increase or Novel	Fold increase or Novel	References
	Leg4	Leg4 + ribose + thiamine 1.5 mM	Leg4 + ribose + thiamine 1.5 mM	Leg4 + ribose + cysteine 1.5 mM	Leg4 + ribose + cysteine 1.5 mM	(Leg4 + ribose + thiamine)	(Leg4 + ribose + cysteine)	
Ketones								
2,3-Butanedione		2,161,257	78,563,251	1,958,985	65,540,109	36	33	Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-Butanone			25,239,728	557,993	12,758,206	Novel	23	Moon et al. (2006, Food Res. Int., 39:294-308); Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85)
2-Heptanone			415,292		371,172	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
3-Octanone			40,094		76,618	Novel	Novel	Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2,3-Pentanedione		67,534	896,415		523,473	13	Novel	www.thegoodscentscompany.com
S-Heterocyclic compounds								
2-Methyl-3-furanthiol			14,315		236,221	Novel	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); www.thegoodscentscompany.com; Mottram (1998, Food Chem., 62:415-24)
2-methyl-Thiazole			25,979		163,919	Novel	Novel	Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
Thiazole			21,109	229,262	10,868,037	Novel	#NAME?	Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
2-Acetylthiazole				14,712	512,095	ND	#NAME?	Song et al. (2012, J. Chromatog. A, 1254:115-24); McGorrin et al. (2001, Special Publication - Royal Soc. Chem., 274:67-84); www.thegoodscentscompany.com; Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8)
Dihydro-3-(2H)-thiophenone					112,049	ND	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); McGorrin et al. (2001, Special Publication - Royal Soc. Chem., 274:67-84); www.thegoodscentscompany.com; Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8)
3-Thiophenemethanol					282,483	ND	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Farmer 1991 cited
3-methyl-Thiophene		217,425	1,376,548		205,936	#NAME?	Novel	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
Thiophene		149,193	683,022	90,079	2,702,671	#NAME?	#NAME?	www.thegoodscentscompany.com; Burdock (2010, Fenaroli's handbook of flavor ingredients, 6th Ed.)
4-methyl-5-Thiazoleethanol			39,748			Novel	ND	www.thegoodscentscompany.com
Sulfides								
dimethyl-Disulfide	15,378	36,281	237,336	33,880	69,922	6	2	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Dimethyl trisulfide		1,264	51,164	24,473	46,195	40	2	Rochat; Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Carbon disulfide		1,000,948	18,591,830	1,209,153	28,040,380	19	23	Jayasena et al. (2013, Trends in Food Sci. & Technol., 34:96-108)
Aldehydes								
2-methyl-Propanal			18,511,383	405,613	18,666,330	Novel	46	Guth & Grosch (1995, Z. Lebensm Unters Forsch, 201:25-6)
Acetaldehyde	2,118,924	1,845,887	67,952,925	5,414,934	51,772,491	36	9	Moon et al. (2006, Food Res. Int., 39:294-308); Guth & Grosch (1995, Z. Lebensm Unters Forsch, 201:25-6)
Pentanal			159,313		381,708	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308)
Benzaldehyde	37,128	86,183	13,953,129	248,601	7,208,946	161	29	Moon et al. (2006, Food Res. Int., 39:294-308)
Butanal					49,139	ND	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Nonanal			231,989		525,261	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Undecanal			361,596	27,895	316,482	Novel	11	Moon et al. (2006, Food Res. Int., 39:294-308); Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
3-methyl-Butanal			22,944,561	406,703	22,645,420	Novel	56	Moon et al. (2006, Food Res. Int., 39:294-308); Rochet & Chaintreau (2005, J. Agric. Food Chem., 53:9578-85)
Furfural			367,866		211,815	Novel	Novel	Resconi et al. (2013, Molecules, 18:6748-81)

Decanal			124,311		107,277	Novel	Novel	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Morita et al. (2003, J. Sci. Food Agric., 83:289-97); Moon et al. (2006, Food Res. Int., 39:294-308); Song et al. (2012, J. Chromatog. A, 1254:115-24); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8); www.thegoodscentscompany.com
3-methyl-2-Butenal			41,782		36,857	Novel	Novel	www.thegoodscentscompany.com; Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-methyl-Butanal		45,898	14,311,236	232,394	7,137,316	312	31	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
Octanal			158,036		124,554	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-Propenal		542,416	1,579,560	459,857	3,596,886	3	8	
Alcohols								
1-Hexanol			107,946		133,744	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-Ethyl-1-hexanol	49,233	56,239	199,266	53,611	169,030	3	2	Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2-Furanmethanol			15,707			Novel	ND	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher); Farmer & Patterson (1991, Food Chem., 40:201-5)
N or O heterocycle								
2-Acetyl-1-pyrroline			19,866		22,489	Novel	Novel	Moon et al. (2006, Food Res. Int., 39:294-308); Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
methyl-Pyrazine			214,505		160,388	Novel	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
2-methyl-Furan			56,185		110,826	Novel	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Kerth et al. (2013, White Paper: Product Quality, Texas A&M University Dept of Animal Sciences); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com
Pyrazine			438,672	18,939	402,961	Novel	21	Song et al. (2012, J. Chromatog. A, 1254:115-24); Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)
2-pentyl-Furan			23,414		21,576	Novel	Novel	Song et al. (2012, J. Chromatog. A, 1254:115-24); Moon et al. (2006, Food Res. Int., 39:294-308); www.thegoodscentscompany.com; Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Karahadian & Lindsay (1989, J. Amer. Oil Chem. Soc., 66:1302-8); Leduc et al. (2012, J. Sci. Food Agric., 92:2560-8)
2,5-dimethyl-Pyrazine			22,739		27,137	Novel	Novel	www.thegoodscentscompany; Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97) ; Van Ba et al. (2012, "Principle of meat aroma flavors and future prospect," Ch 7 in Latest Research into Quality Control, InTech Open Access Publisher)
2,3-dimethyl-Pyrazine			25,077		34,400	Novel	Novel	www.thegoodscentscompany.com; Brewer (2006, "The Chemistry of Beef Flavor – Executive Summary," National Cattlemen's Beef Association, #12814); Morita et al. (2003, J. Sci. Food Agric., 83:289-97)

Attorney's Docket No.: 38767-0015001 / M3CON-Track 1

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

First Named Inventor	:	Rachel Fraser	Art Unit	: 1791
Serial No.	:	14/797,006	Examiner	: Elizabeth A. Gwartney
Filed	:	July 10, 2015	Conf. No.	: 8496
Title	:	METHODS AND COMPOSITIONS FOR AFFECTING THE FLAVOR AND AROMA PROFILE OF CONSUMABLES		

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 35 U.S.C. § 1.132

I, Keith R. Cadwallader, Ph.D., hereby declare as follows:

1. I am currently a Professor of Food Chemistry in the Department of Food Science and Human Nutrition at the University of Illinois in Urbana, Illinois. I have held this position since 2007. I have worked and taught in the fields of Food Science, Food Chemistry and Flavor Science since 1985. Prior to my current position, I held Assistant/Associate Professor positions in Food Chemistry at Louisiana State University and Mississippi State University and was an Associate Professor at the University of Illinois.

2. I received my B.S. degree in Food Science from the University of Georgia in 1985, my M.S. degree in Food Science from the University of Florida in 1987, and my Ph.D. degree in Food Science (minor in Chemistry) from the University of Florida in 1990.

3. A copy of my curriculum vitae is attached as **Exhibit A**.

4. My research includes: chemistry and analysis of food and beverage flavors, flavor (aroma) characterization and flavor quality determination, instrumental and sensory identification and quantification of character-impact flavor compounds in foods/beverages and agricultural/ industrial materials, process flavor development, recovery of flavor constituents

from food processing by-products, development of methods to stabilize labile flavors, occurrence and mechanisms of flavor formulation in foods and beverages, technology for production of food and beverage flavors, and physical chemistry of food/beverage matrix-flavor binding, flavor release and flavor-packaging interactions. I have taught courses relating to food analysis, flavor chemistry, food products analysis, applied food chemistry, flavor and food acceptance, food composition, and flavor/color of foods.

5. I am a nationally and internationally recognized expert and scholar in the field of flavor science. I have co-edited five books and published 57 refereed book chapters and 96 refereed journal articles in the area of food flavor chemistry and analysis. My research has been published in well-respected journals in the field of flavor chemistry, including the *Journal of Agricultural and Food Chemistry*, *Food Chemistry*, *Journal of Food Science*, *Analytical Chemistry*, *Journal of Sensory Studies*, *Journal of the American Oil Chemists Society*, *Cereal Chemistry*, *Journal of Dairy Science*, *Flavour and Fragrance Journal* and others. I have presented more than 50 invited symposium talks and over 115 scientific papers at national and international scientific meetings. I am co-inventor of an invention described in a U.S. patent application related to stabilized compositions and methods of manufacture (Publication No. US 2014/0275543 A1).

6. Through my research and teaching, I have made both fundamental and applied contributions to the current understanding of flavor chemistry and analysis. In 2016, I received the University of Illinois, College of Agriculture, Consumer and Environmental Sciences, *Senior Faculty Award for Excellence in Research.* I was awarded the *2007 Stephen S. Chang Award for Excellence in Lipid or Flavor Science* from the Institute of Food Technologists. In 2010, I was

elected a *Fellow in the Agricultural and Food Chemistry Division of the American Chemical Society* in recognition of my scientific contributions to the field of agricultural and food chemistry, specifically in the area of flavor science.

7. I am a professional member of the Institute of Food Technologists and the American Chemical Society (Agricultural and Food Chemistry Division). From 2003-2005, I was the Chair and Program Chair of the Agricultural and Food Chemistry Division of the American Chemical Society. I currently serve on the editorial board of the *Flavour and Fragrance Journal* and as an associate editor of the *Journal of Food Science*.

8. In the preparation of my declaration, I have reviewed the U.S. Application Serial No. 14/797,006 ("the '006 application"), as well as the Office Action mailed June 6, 2016, in which the claims of the '006 application were rejected as being anticipated by Proulx et al. (2006, J. Agric. Food Chem., 54:1518-22) ("Proulx") and as being obvious considering Hsieh et al. (US 2012/0093994) ("the '994 application") in view of Proulx. I have also considered additional materials listed in **Exhibit B**.

9. Flavor is the integrated response to the simultaneous perception (total or combined sensation) of the taste, aroma and somatosensory (nerve response) characteristics of substances naturally present in foods. Although the peripheral sensory organs for detection of taste and smell stimuli are quite distinct, their signals are integrated in the orbitofrontal and other areas of the cerebral cortex of the brain to generate the perception of "flavor" and to mediate food recognition and overall acceptance (Chaudhari and Roper, 2010). It is generally accepted by most researchers that olfaction (aroma) plays the dominant role in the flavor of food (Spence, 2015), and aroma has been regarded as the single most important factor contributing to the

characteristic flavor of meat (Shahidi et al., 1986).

10. Gas chromatography-mass spectrometry (GC-MS) has been the method of choice for the analysis of aroma components of meat for over five decades (Cadwallader and MacLeod, 1998). Before GC-MS can be conducted, the volatile components of the product must first be isolated from the sample and then transferred to the instrument for subsequent analysis. Headspace solid phase microextraction (HS-SPME) combined with GC-MS has been widely and successfully used for analysis of aroma components of meat (Rochat et al., 2007) and "meat aroma" reaction mixtures (Jublot et al., 2010). During analysis, the GC-MS system separates the individual volatile compounds in the mixture and then the mass-spectrometer detects and characterizes each component individually, enabling both qualitative (compound identification) and quantitative (compound concentration) analyses to be performed.

11. In my opinion, Applicants discovered a "ground beef-like food product" containing no animal products and including:

0.01%-5% by weight of a heme-containing protein comprising an amino acid sequence having at least 80% sequence identity to the polypeptide set forth in SEQ ID NO:4;

a sugar, sugar alcohol, sugar acid, or sugar derivative selected from glucose, ribose, fructose, lactose, xylose, arabinose, glucose-6-phosphate, maltose, and galactose, and mixtures of two or more thereof; and

at least 10 mM of a compound selected from cysteine, cystine, selenocysteine, thiamine, methionine, and mixtures of two or more thereof; and

at least 10% or more by weight one or more plant proteins,

which has the surprising capability of promoting, upon heating, the efficient conversion of

nonvolatile flavor precursors into desirable meaty aroma compounds. This is a significant discovery since a person having ordinary skill in the art would not have anticipated that the inclusion of a heme protein would have any beneficial effect on flavor. In fact, iron, and specifically heme iron, has been most often associated with causing deleterious off-odor consequences in foods and especially meat products (Calkins, 2007). As a flavor scientist who has worked in the field for over 30 years, I consider Applicants' discovery to be completely unexpected.

12. In forming my opinion, I reviewed and considered Applicants' experiments related to their discovery and demonstration that the addition of heme protein (in some cases as leghemoglobin or LegH) is required or essential for the development of meaty aroma in a "ground beef-like food product" comprising no animal products. These experiments are described in the '006 application. In addition, I reviewed the experiments in the Declaration Under 37 CFR §1.132 of Dr. Celeste Holz-Schietinger, Ph.D. ("Holz-Schietinger Declaration").

13. The Hotz-Schietinger Declaration describes the application of state-of-the-art instrumental aroma analysis methods, specifically the aforementioned HS-SPME combined with GC-MS, to monitor the effect of LegH addition on the formation of volatile compounds, specifically those with meaty aroma characteristics, during heating (i.e., cooking). The objective was to demonstrate that LegH was capable of promoting or catalyzing the formation of meaty aroma compounds via the Maillard reaction and other thermal reactions between ribose and cysteine. Three flavor reaction systems were compared, consisting of: 1) 1% (10 mg/mL) LegH alone (**LegH control**), 2) 50 mM ribose + 50 mM cysteine alone (**Rib + Cyst**) and 3) 1% (10 mg/mL) LegH + 50 mM ribose + 50 mM cysteine (**LegH + Rib + Cyst**). The above reaction

mixtures were diluted in a buffer (50 mM potassium phosphate and 100 mM sodium chloride) and adjusted to pH 6.5, which is near the physiological pH of ground beef. A 2 mL aliquot of each reaction mixture was transferred to sample vials (sealed) which were subsequently heated (150C for 3 min) and then analyzed by HS-SPME-GC-MS.

14. It is my opinion that the above experimental approach and subsequent data analyses techniques used by Dr Celeste Holz-Schietinger are appropriate and acceptable for the purpose of evaluating and comparing aroma compound formation in the three reaction systems under heating conditions. It is also my opinion that these conditions which would simulate the cooking of a "ground beef-like food product" containing no animal products as described in the '006 application, and that the data obtained from the experimental approach would be reasonably predictive of the expected aroma compound formation from a "ground beef-like food product" containing no animal products and including the flavor reaction system components LegH, ribose, and cysteine.

15. In my opinion, Applicants' experiments in the '006 application clearly demonstrate that inclusion of LegH in the reaction mixture has the surprising result of greatly enhancing the reaction of ribose and cysteine to form a variety of volatile compounds possessing meaty aromas. Moreover, it is my opinion that Dr. Celeste Holz-Schietinger's experiments confirm these surprising results. Among the 28 compounds identified in her experiments, two in particular are well-known to possess meaty sensory characteristics and to be important and potent odorants in cooked meat. Specifically, 2-methyl-3-furanthiol (MFT) and its dimerized form bis(2-methyl-3-furyl)disulfide (MFT-MFT) are important aroma compounds in cooked meat (Tang et al., 2013). Both compounds have extremely low odor detection thresholds of 0.7

parts-per-trillion (pptr) and 0.02 pptr, respectively (Belitz et al. 2006), and are usually only present in low abundance (pptr range) in cooked meat (Tang et al., 2013). Despite their low abundance in meat they are still among the most impactful aroma compounds because of their extremely low odor detection thresholds.

16. On the basis of my considerable knowledge and experience in the field of flavor science, it is my opinion that Applicants have indeed discovered and demonstrated the unique ability of heme-protein, including LegH, to serve as an efficient catalyst or promoter of the Maillard reaction and/or other relevant thermal reactions (e.g. thiamine degradation) which can form meaty aroma compounds in a "ground beef-like food product" containing no animal products.

17. It is also my opinion that a person having ordinary skill in the art would not have anticipated or expected this discovery based on the prior art cited. My opinion takes into consideration the Proulx et al. (2006) study, which did not disclose, mention or otherwise anticipate the potential for a heme-protein to influence or promote the formation of desirable meaty aroma compounds. Furthermore, Applicants' unique flavor precursor system, in which a heme-protein is intentionally added to act as an essential promotor of meat flavor generation during cooking, was not disclosed, mentioned or otherwise anticipated in the '994 application. My opinion also takes into consideration the common understanding of those in the field that the inclusion of iron, including heme iron, has been most often associated with causing deleterious off-odor consequences in foods and especially meat products.

18. I hereby declare that all statements made herein of my knowledge are true and that all statements made on information and belief are believed to be true; and further that these