THESIS

PROFILING AND PUTATIVE AROMA BIOMARKER IDENTIFICATION FOR FLAVOR IN POTATOES USING A TRAINED SENSORY PANEL AND HS-SPME GC-MS

Submitted by

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In partial fulfillment of the requirements

For the Degree of Master of Science

Colorado State University

Fort Collins, Colorado

Spring 2017

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ABSTRACT

PROFILING AND PUTATIVE AROMA BIOMARKER IDENTIFICATION FOR FLAVOR IN POTATOES USING A TRAINED SENSORY PANEL AND HS-SPME GC-MS

Flavor is the synthesis of taste and aroma sensations. The taste fraction of flavor, including salty, sweet, sour, bitter, and savory, refers to non-volatile chemical compounds that are detected by epithelial cells in the mouth. The aroma fraction encompasses volatile or semi-volatile chemical compounds that are sensed by nerve cells within the olfactory system, particularly during the chewing process, initiating a multitude of sensations such as earthy, floral, or fruity. Flavor may also be influenced by texture, sound, appearance, or personal preference, resulting in an inherently complex phenotypic trait that is difficult to assess. Cooked flavor profiling of fifteen fresh market potato clones (*Solanum tuberosum,* L.) using sensory analysis paired to aroma compound analysis reveals potential biomarkers for flavor phenotyping. Trained sensory panelists described extensive, significant sensory differences between potatoes including bitter, buttery, creamy, earthy, fruity, off-flavors other than bitter, potato-like flavor, sweet, woody, appearance, aroma intensity, mealy texture, and overall quality (mixed model ANOVA, α=0.05, n=17-38 ratings x 15 clones). Non-targeted volatile metabolomics with headspace solid-phase microextraction gas chromatography coupled to mass spectrometry (HS-SPME GC-MS) facilitated identification of 42 unique metabolites with significant variation across samples (ANOVA, α=0.05, n=5 technical replicates x 14-15 clones x 2 cooking methods). Based on Spearman's rank correlations, hierarchical clustering analysis (HCA), and principal component analysis (PCA), potential biomarkers for buttery, a positive flavor attribute, are aldehydes 1-nonanal, benzaldehyde, (E)-2-heptenal, pentanal, 2-phenylacetaldehyde, the alcohol (Z)-2-

methyl-2-penten-1-ol, and 5-methyl-2-hexanone, a ketone. Other positive flavor attributes, sweet and potato-like flavor, are related to benzoate-3-methyl-2-buten-1-ol, 2-ethylfuran, and 3,4,5-trimethyl-2-cyclopenten-1-one. Potential biomarkers for negative flavor attributes are also implicated. Additionally, some sensory and aroma compound differences occur between baked versus boiled potatoes. Flavor biomarkers may play a key role in achieving flavor quality improvement through breeding and selection.

ACKNOWLEDGEMENTS

The Colorado Potato Administrative Committee and the United States Department of Agriculture Specialty Crop Grant Initiative graciously funded this project. First and foremost, thank you to David and Sastry for bolstering my experience in field and lab work, as well as patience and mentorship during the process. Of course I am thankful for the commitment of sensory panelists over two years, but they were also kind, interesting, and enjoyable people. I'm also greatly appreciative of the training, assistance, and friendship from Caroline Gray, Mike Gray, and Katrina Zsavislan in the San Luis Valley. Thank you to Sharon Yust and Michelle Leckler for their support, answering questions, helping me locate supplies, and to Sharon especially for always having a full candy bowl. For their consistently positive attitudes and peer support, thank you to Diganta Kalita, Mansour Hamed, and Esam Emragi. Thank you to Adam Heuberger and Martha Stone for helpful technical advice in metabolomics and sensory analysis, respectively. The Department of Horticulture & Landscape Architecture office staff on campus was always warm, patient, and helpful as well. I'm extremely grateful to have met Ryan Fulgham during my Master's work, and his unconditional encouragement, companionship, and ability to keep me mostly sane. Many thanks to Sandra Fortson, Jasmine Klauder, Steve Book, Jacob Vigil, and Colin Wakeham too for forming my Colorado family. My dog, Effie, has also been an important part of my life the past few years, keeping me mentally and physically fit despite occasional graduate student blues. Finally, thank you to Monster Energy for keeping me awake during long drives and work weeks.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1. LITERATURE REVIEW

1.1. Introduction

Potato, a significant national and global food staple, is a tuber crop originating in the Andean Mountains of South America. Despite the bland flavor of the most commonly cultivated potatoes, potatoes contain considerable flavor diversity in terms of taste, aroma, and texture. An assortment of shape, size, skin, and flesh colors exists as well. Although flavor is a complex and subjective trait, it has been examined through extensive metabolite analyses with some sensory analysis correspondence in potato. Nonetheless, potato metabolite corroboration with human flavor sensations is relatively lacking but crucial to fresh market crop improvement through breeding and selection.

1.2. Potato As a Crop

1.2.1. General Taxonomy and Characteristics

Potatoes, potato tubers, or simply tubers are modified underground stems that store nutrients and enable asexual propagation. Within the nightshade family, Solanaceae, potatoes belong to the genus *Solanum,* which includes both wild and cultivated potato relatives.[1] The most commonly cultivated potato species is tetraploid (2n=48) *Solanum tuberosum* (L.).[1,2] Potatoes are up to 79% water yet satisfy daily human dietary energy and protein requirements with an ample supply of B and C vitamins, calcium, iron, phosphorous, and potassium.[3]

1.2.2. Origin and Early Cultivation

Though wild species of *Solanum* range from the central United States to south central Chile[1], the center of origin of all *Solanum* species is most likely the Andean Mountains in Peru and Bolivia.[1,2,4] Archaeological evidence confirms that potatoes were cultivated at least as early as the second century in South America, but there is speculation that potatoes may have been cultivated up to 6,000 to 8,000 years ago.[2,4] In the late sixteenth century, the import of potatoes to Europe prompted worldwide dissemination of potatoes as a staple food crop.[2,5]

1.2.3. Current Cultivation

Today, potatoes remain in the top rankings of worldwide staple food crops in terms of production acreage, dietary intake, and economic livelihood. Each year, the United States National Potato Council trade association compiles data from the United States' Departments of Agriculture and Commerce as well as the Food and Agriculture Organization of the United Nations into a *Potato Statistical Yearbook*. The most recent issue, the *2016 Potato Statistical Yearbook*, indicates that the United States was the 5th largest global potato producer in tons for 2015, of which 24% of potatoes were sold to the fresh market sector and 67% for processing.[6] For the same year, total production value in the United States was nearly $4 billion. Nationally, Colorado ranked as the 5th largest producer in tons. Readers are encouraged to refer to the most recent issue of the *Potato Statistical Yearbook* as it becomes available.

1.3. Potato Flavor

1.3.1. Overview of Flavor

Flavor is primarily a combination of sensations produced by taste and aroma receptors on the tongue or in the olfactory, respectively.[7] The taste fraction of flavor refers to non-volatile metabolites, whereas the aroma fraction encompasses volatile metabolites.[8] Volatiles are substances that readily vaporize from solid or liquid to gaseous forms. Contrastingly, non-volatiles remain in a solid or liquid form without chemical modifications or extreme physical conditions. Flavor may also be influenced by texture, appearance, and sound.[9] In potatoes, texture, or mouthfeel, has the most predominant influence on overall flavor after taste and aroma.[10]

1.3.2. Taste

Taste sensations are more readily identifiable than aromatic sensations, including sweet, salty, sour, bitter, and umami. Modified epithelial cells on the tongue and in the mouth confer taste through the reception of non-volatile compounds.[7] Most early potato flavor research from the 1950s to 1970s focuses on bitter taste compounds, specifically glycoalkaloids and phenolics, which are the main contributors to off-flavors, particularly bitterness and astringency.[3,11–13] The amino acids leucine, isoleucine, and phenylalanine lysine also contribute to bitterness.[14] Although sweet, salty, and sour compounds are not thought to have a noticeable influence on potato flavor, one sensory analysis study indicated a positive correlation between perceived quality and sweetness.[15] Umami taste is more prominent in potatoes,[16,17] resulting from the interaction between certain amino acids and 5'-nucleotides released during cooking.[18] Multiple studies[11–13,19–22] show that total amounts of potato taste compounds vary by cultivar, environment (e.g. location and year), and even storage temperatures.[14]

1.3.3. Aroma

Flavor diversity beyond the five basic taste components is primarily ascribed to aroma compounds, which may or may not be aromatic, including semi-volatile and volatile compounds. Volatiles and semi-volatiles are sensed by olfactory nerve receptors through smell and the act of eating, the latter of which causes air to pass backwards through the nasal cavity.[7] As suggested by the amount of research literature for aroma versus taste compounds in potato, aroma influences overall flavor to a greater degree than taste. One study indicated that a lack of volatiles resulted in decreased detectable flavor evaluated by a sensory panel.[23] Most aroma metabolites related to potato flavor result from interactions during cooking between fatty acid, sugar, and amino acid precursors, including Strecker amino acid degradation to form aldehydes, lipid degredation to aldehydes and ketones, Maillard reactions that form pyrazines, and other sugar degradation reactions.[24,25] Sulfur metabolites are also important to potato flavor,[25,26] as are terpenes and pyrazines present in raw tubers.[10] Relative importance of certain aroma compound types to human potato flavor sensations is debated. Some researchers attest that pyrazines are most significant,[27–29] whereas others suggest that aldehydes are more important,[25,30] though most researchers agree that a mixture of several aroma compounds rather than a singular constituent imparts potato flavor.

Hundreds of volatile or semi-volatile metabolites have been associated with potato flavor. As with taste metabolites, aroma metabolites differ by cultivar,[31–33] environment,[32] and storage conditions.[25,30] However, unlike taste metabolites, potato aroma metabolites vary by cooking method.[33,34] Though most aroma studies utilize a single cooking method, one study directly compares cooking methods and demonstrates that baking or microwaving potatoes results in drier flesh that favors Maillard reaction and sugar degradation products, whereas boiling enables increased accumulation of terpenes and pyrazines.[33] Another study indicates that although volatiles differ among select Italian potatoes, geographical origin cannot be distinguished based on volatiles alone.[35]

1.3.4. Texture

Desirable potato texture depends on cooking or processing utilization. Texture characteristic thresholds are more critical for potatoes that will be used for chips, fries, dehydrated products, or other food products and will not be discussed within this thesis. Texture attributes for fresh market potatoes are less defined, but can be described as mealy or waxy.[10]

Mealy potatoes are dry, with more starch, and a greater specific gravity compared to waxy potatoes, which are gummier, have less starch, and a smaller specific gravity.[10,36] Red and yellow flesh potatoes tend to be waxy, whereas white flesh potatoes tend to be mealy.[36] In a sensory analysis that includes mealiness for several potato cultivars, mealiness is not related to overall quality perception and is variable across cultivars and environment.[15]

1.4. Methods for Evaluating Potato Flavor

1.4.1. Flavor Compound Analysis

Taste components of potato flavor, including glycoalkaloids, sugars, and amino acids are typically detected by mass spectrometry (MS), ultraviolet detection (UV), or flame ionization detection (FID) and quantified by coupling to high-performance liquid chromatography (HPLC). An enzyme-linked immunosorbent assay (ELISA) has also been developed for glycoalkaloids specific to potatoes.[20] Still, assays for taste are fairly laborious, though efforts have been made to shorten extraction times.[37] For aroma compound measurement, gas chromatography (GC) is typically paired to MS for volatile identification and quantification or an olfactometry FID sniffing port for aroma characterization. A multitude of extraction methods causes aroma compound methods to be more variable than for taste, but can be separated into liquid or headspace (HS) methods.

In liquid extraction methods, aroma compounds are extracted with solvents. Simultaneous-distillation extraction (SDE) is an older liquid extraction method that tends to be tedious, to require large quantities of potatoes, and to result in numerous metabolite artifacts.[23] Solvent-assisted flavor evaporation (SAFE) is similar to SDE but results in fewer artifacts.[31,32,38,39] Newer liquid extraction methods incorporate derivitization, the chemical modification of aroma compounds from volatiles to liquids, which has less extraction bias, requires less sample material, and is more reproducible than SDE or methods.[21,22,40,41]

With solid-phase polymers rather than solvents, aroma compounds are extracted via adsorption in HS extraction. Though early HS extraction in potatoes required a large amount of potatoes,[29] the development of Tenax plate dynamic HS concentration methods greatly reduced sample size requirements and increased reproducibility.[25,30,33] Solid-phase microextraction (SPME) from HS is comparable to dynamic HS method volatile profiles but with slightly less sensitivity, greater automation capability, and faster extraction times.[35,42–45]

1.4.2. Sensory Analysis

The subjective nature of flavor requires human sensory analysis. Even if potato cultivars exhibit unique chemical profiles, sensory analysis is necessary to determine if differences in chemical profiles can be detected.[33] Several sensory methods exist, but the most common approach for potato flavor evaluation is a trained panel for descriptive profiling. Trained panelists may also be used for aroma characterization via GC olfactory sniffing ports. Training methods are poorly described, while scorecards and attributes are inconsistent. Some sensory scorecards use a non-graded linear rating scale,[15,16,31,45,46] others use a categorical hedonic scale.[11,13,38,44] Examples of detectable potato attributes are sweetness, flavor intensity, bitterness, astringency, umami, and mealiness.[11,13,15,45] Sensory analysis is resource-intensive, which may explain the limited number of described potato attributes.[7]

1.4.3. Instrumental Texture Analysis

Though some sensory panels have evaluated cooked potato texture, instrumental texture analysis is largely unexplored. For most foods, however, single variable instrumental measurements typically do not correlate to sensory texture ratings.[7]

1.4.4. Integrated Methods

As already mentioned, sensory analysis is crucial in identifying flavor compounds that influence potato flavor. Several important flavor compounds have been identified by combining sensory and chemical methods,[16,44,45] which has the potential to reveal key flavor metabolic pathways and genes with additional molecular techniques. One such combination demonstrates that potato off-flavor and some aldehyde flavor compounds associate with lipoxidase activity.[44] Specific genes have also been associated with potato flavor.[43]

1.5. Conclusion

Plant breeding efforts tend to focus on yield, postharvest storage, and pest resistance, which is thought to have a negative influence on produce quality, including flavor.[4,7] Though past aroma compound extraction methods are slow and resource-intensive, SPME provides a fast, solvent free method that enables more efficient metabolic phenotyping.[41] Flavor gene discovery coupled with efficient phenotyping may play a vital role in flavor crop improvement.[47] Initial sensory analysis should be used to identify critical flavor compounds prior to potential gene discovery.

The experiments described in the remaining thesis explore potato flavor through sensory, metabolite, and instrumental texture analysis of select potato clones. Overall objectives were as follows:

1) to generate and compare cooked flavor profiles of potato cultivars

2) to identify volatile and semi-volatile flavor metabolites of cooked potatoes potato

3) to evaluate instrumental texture analysis of cooked potatoes

Unlike other potato flavor studies that contain a sensory component, the following sensory analysis covers an extensive array of flavor attributes that are used to identify significant flavor metabolites. These flavor metabolites provide a framework for future potato flavor gene discovery studies. Further development of metabolite detection methods may provide a means of high-throughput flavor phenotyping, which would be particularly useful to selection-based breeding programs such as the Colorado State University Potato Breeding and Selection Program. Flavor profiles generated from sensory analysis are also informative to potato breeders, producers, and consumers that wish to maximize flavor quality.

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CHAPTER 2. PLANT MATERIAL AND GENERAL PREPARATION

2.1. Introduction

Potato tuber material from two growing seasons at the San Luis Valley Research Center (SLRVC), a Colorado State University (CSU) Agriculture Experiment Station (AES) located in Center, CO was analyzed for sensory, volatile compound, and texture components. Fifteen clones, including six advanced selections and nine named cultivars with a variety of flesh and skin colors were evaluated. Potatoes were baked or boiled then cut and served warm during sensory sessions. After sensory sessions, remaining warm potatoes were used for texture analysis or frozen with liquid N_2 and stored at -80°C until volatile analysis.

2.2. Clone Characteristics

Potato clones consisted of commercial cultivars and advanced selections from the Colorado Potato Breeding and Selection Program at the SLVRC. White-fleshed clones (all russets) included named cultivars Russet Norkotah, Russet Burbank, Fortress Russet, Russet Nugget, and advanced selection AC00395-2RU. Yellow-fleshed clones encompassed named cultivars Masquerade, Harvest Moon, Red Luna, Yukon Gold, and advanced selections CO04067-8R/Y and CO04099-3W/Y. Red- and purple-fleshed clones were named cultivar Purple Majesty and recently named Crimson King as well as advanced selections CO04056-3P/PW and CO04063-4R/R. In addition to flesh color, clones had various skin colors, size classes, and origins (Table 2.1).

2.3. Cultural Conditions

All potato clones were field grown in 2014 and 2015 by the Colorado Potato Breeding and Selection Program at the SLVRC, with the exception of advanced selection CO04063-4R/R, which was grown in 2014 only. Soil type is a sandy loam classified as Dunul cobbly sandy loam. Plots were spaced 86 cm apart with a length of 7.6 m by 25 hills with 30 cm spacing between hills. Planting, vine kill, harvesting dates and number of days grown from planting until vine kill were similar for both years (Table 2.2). Irrigation was provided as necessary by evapotranspiration using a center pivot at a 42 and 38 cm gross application rate in 2014 and 2015, respectively. Rainfall during the growing season was 3.6 cm in 2014 and 12.0 cm in 2015. Fertilization rates were the same for both years (Table 2.3) while pesticide applications were somewhat different (Table 2.4). In 2014, vine-kill was accomplished with Reglone (Syngenta AG, Basel, Switzerland) at a rate of 9 L per ha. Sulfuric acid at a rate of 38 L per ha was used for vine-kill in 2015. Harvested tubers were stored at approximately 4.4 °C with a relative humidity of 95% for at least 3 months.

2.4. Tuber Preparation

Potato tubers free of imperfections from each clone were used for sensory, volatile, and texture analysis. Preparation of tubers prior to cooking involved a combination of tap water rinsing and gentle scrubbing. Tubers intended for baking were punctured with a fork, wrapped in aluminum foil, and cooked until easily penetrated with a toothpick at 200 °C (45 minutes to an hour and a half, varying by clone). Boiled tubers were cooked in separate pots of water by clone brought to a boil before the addition of tubers. Large-sized tubers were cut in half for boiling. Sensory analysis samples included six 1 cm cooked potato cubes, approximately two from a single tuber of a clone. At least three different tubers were used for sensory samples. Small size class tubers generally required one or two more tubers. Sensory panelists received warm, cooked potatoes in 8 oz Styrofoam cups covered with aluminum foil and coded with randomized 3-digit codes by clone. For volatile analysis samples, about 75 g of finely diced cooked potatoes from both tuber stem and bud ends were bulked from at least three different tubers, frozen with liquid nitrogen, and stored at -80 °C until analysis. All sensory and volatile analysis samples were free of tuber skin. The texture of at least eight whole cooked tubers per clone was analyzed for 2014, whereas 30 tubers from each clone were used for 2015.

2.5. Summary

Analysis included a diverse array of potato clones that were field grown using standard practices for the San Luis Valley region in Colorado. Potato tubers were prepared similarly for sensory, texture, and volatile analyses through either baking or boiling cooking methods. Growing conditions were similar for both years that tubers were harvested, though more precipitation occurred in 2015 versus 2014.

Table 2.1. Characteristics of field grown potato clones evaluated for flavor[a] at the SLVRC (Center, CO)

Clone	Code	Skin Color	Flesh Color	Size[b]	Origin[c]
AC00395-2RU	95	Russet	White	Large	A95523-12 x Summit Russet
CO04056-3P/PW	56	Purple	Purple	Small	CO97216-1P/PW x CO97277-2P/PW
CO04063-4R/R[d]	63	Red	Red	Medium	CO97226-1R/R x CO97222-1R/R
CO04067-8R/Y	67	Red	Yellow	Medium	CO97232-1R/Y x ATC98444-1R/Y
CO04099-3W/Y	99	White	Yellow	Medium	VC1002-3W/Y x ATC98495-1W/Y
Crimson King	CK	Red	Red	Medium	CO94170-1 x Mountain Rose
Fortress Russet	FR	Russet	White	Large	AC99375-1RU: AWN86514-2 x A89384-10
Harvest Moon	HM	Purple	Yellow	Small	AC99330-1P/Y: Inka Gold x A89655-5DY
Masquerade	MQ	Purple and White	Yellow	Medium	AC99329-7PW/Y: Inka Gold x A91846-5R
Russet Norkotah	NK	Russet	White	Large	ND9526-4 Russ x ND9687-5 Russ
Purple Majesty	PM	Purple	Purple	Medium	CO94165-3P/P: ND2008-2 x All Blue
Red Luna	RL	Red	Yellow	Medium	CO97233-3R/Y: CO94218-1 x VC0967-5
Russet Burbank	RB	Russet	White	Large	
Russet Nugget	RN	Russet	White	Large	Krantz x AND71609-1
Yukon Gold	YG	White	Yellow	Medium	Norgleam x W5279-4

[a]Potato clones were grown in 2014 and 2015 but evaluated in 2015 and 2016, respectively.

[b]Size class according to USDA United States Standards for Grading of Potatoes, where small tubers have a diameter of 4.45 to 6.35 cm (1.75 to 2.50 in), medium tubers have a diameter of 5.72 to 8.26 cm (2.25 to 3.25 in), and large tubers have a diameter of 7.62 to 11.43 cm (3.00 to 4.50 in).

[c]Origin refers to the parents that were crossed to produce the progeny from which clones were propagated.

[d]Grown in 2014 only.

Table 2.2. Growing season of potatoes evaluated for flavor[a] at the SLVRC (Center, CO)

	2014	2015
Planting Date	16-May	15-May
Vine Kill Date	4-Sep	2-Sep
Harvesting Date	29-Sep	29-Sep
Number of Days Grown	108	110

[a]Potato clones were grown in 2014 and 2015 but evaluated in 2015 and 2016, respectively.

Table 2.3. Fertilization schedule of potatoes evaluated for flavor[a] at the SLVRC (Center, CO)

2014	2015	Type	Nutrient	Amount (kg ha^{-1})
16-May	15-May	in-row liquid	N	435
			P_2O_5	326
			K_2O	218
			S	136
			Zn	13
20-Jul	23-Jul	fertigation	N	82
26-Jul	27-Jul	fertigation	N	82
4-Aug	18-Jul	fertigation	N	54
			total N	*653*

[a]Potato clones were grown in 2014 and 2015 but evaluated in 2015 and 2016, respectively.

Table 2.4. Pesticide schedule for potatoes evaluated for flavor[a] at the SLVRC (Center, CO)

	2014	Product	Amount a.i. (kg ha^{-1})	2015	Product	Amount a.i. (kg ha^{-1})
Insecticide	17-Jul	Leverage 360[c] (imidacloprid and B-cyfluthrin)	1.0	15-Jul	Leverage 360 (imidacloprid and B-cyfluthrin)	1.0
	24-Jul	Belay[d] (clothianidin)	1.0	4-Aug	Belay (clothianidin)	1.0
	8-Aug	Movento[c] (spirotetramat)	1.7			
Fungicide	12-Jul	Quadris[e] (azoxystrobin)	1.1	15-Jul	Quadris Opti[e] (azoxystrobin and chlorothalonil)	1.1
	24-Jul	Luna Tranquility[c] (fluoryram and pyrimethanil)	3.8	4-Aug	Luna Tranquility (fluoryram and pyrimethanil)	3.8
Herbicide	2-Jun	Outlook[f] (dimethenamide-P)	0.9	5-Jun	Dual Magnum[e]	7.8
	24-Jun	Eptam[g] (S-ethyl dipropylthiocarbamate)	1.0	5-Jun	Boundary[e] (metolachlor and metribuzin)	5.4 and 1.3
	24-Jun	Matrix SG[h] (rimsulfuron)	0.1			

[a]Potato clones were grown in 2014 and 2015 but evaluated in 2015 and 2016, respectively.
[b]The acronym a.i. is synonymous with active ingredient.
[c]Formulated by Bayer CropScience, Monheim am Rhein, Germany
[d]Formulated by Valent Agricultural Products, Walnut Creek, CA
[e]Formulated by Syngenta AG, Basel, Switzerland
[f]Formulated by BASF Crop Protection, Research Triangle Park, NC
[g]Formulated by Gowan Co., Yuma, AZ
[h]Formulated by DuPont, Wilmington, DE

CHAPTER 3. SENSORY ANALYSIS

3.1. Introduction

A number of investigations describe and propose cooked potato flavor metabolite profiles based upon instrumental measurements, however, relatively few flavor profiles created from sensory profiling are defined. Flavor is a subjective, complex quality trait that primarily encompasses taste and olfactory sensations, although appearance, texture, or other characteristics may influence perception.[1] Early potato sensory analysis focuses on off-flavor sensory attributes, including bitterness and astringency.[2,3] More recent profiles include sweetness,[4-6] umami,[7] mealiness,[4] and overall flavor intensity,[5-7] as well as other descriptive attributes.[5,7,8] Moreover, most recent potato sensory studies focus on European cultivars,[5-8] whereas American cultivar descriptions are comparatively absent.

The following study describes sensory-derived flavor profiles of fifteen American potato clones developed from a sensory panel. A trained rather than untrained sensory panel enabled comprehensive profiling for fifteen attributes. Positive and negative attributes are identified using associations to overall quality scores. In addition to clone differences, differences between cooking methods, baking or boiling, and interactions with clone are explored.

3.2. Materials and Methods

3.2.1. Tuber Material

Clones consisted of white-, yellow-, purple-, and red-fleshed named cultivars or advanced selections grown by the Colorado State University (CSU) Potato Breeding and Selection Program at the San Luis Valley Research Center (SLVRC, Center, CO) in 2014 and 2015 for sensory analysis in 2015 and 2016, respectively. White-fleshed clones, all russets, were

AC00395-2RU, Fortress Russet, Russet Norkotah, Russet Burbank, and Russet Nugget. Yellow-fleshed clones were CO04067-8R/Y, CO04099-3W/Y, Masquerade, Harvest Moon, Red Luna, and Yukon Gold. Purple-fleshed clones were CO04056-3P/PW and Purple Majesty, whereas red-fleshed clones were CO04063-4R/R and Crimson King. Tubers were baked or boiled, diced into cubes, and served warm. Prior to baking, tubers were poked with a fork and wrapped in aluminum foil. Randomized material from at least three tubers per clone per cooking method was provided to each panelist. Presentation of coded samples was randomized across panelists per session to account for order effects.

3.2.2. Sensory Analysis

A trained panel of 15 sensory panelists evaluated potato clones, with at least eight panelists present per sensory session. Panelists were recruited as volunteers in the San Luis Valley of Colorado. Demographics were 47% men and 53% women; 80% white, 20% other races; 20% aged 18 to 25, 27% aged 26 to 40, 27% aged 41 to 55, and 27% aged 56 to 70. A preliminary training session for each year involved an overview of tasting technique as well as aroma and taste discussions for reference foods. In this case, sampling technique refers to the inhalation and exhalation of air while chewing to maximize flavor components and the utilization of water and crackers as a palate-cleanser between samples. Reference foods were selected during the training session of the first year and duplicated the second year for flavor attributes (Table 3.1). Following panelist consensus, a scorecard of 15 attributes were listed in the following order: appearance, aroma intensity, potato-like flavor, sweet, fruity, lemon, umami, buttery, creamy, earthy, woody, bitter, off-flavors other than bitter, mealy texture, and overall quality (Appendix 1). Attributes were rated on a scale from 1 to 9 with both numeric and hedonic anchors, from absence to maximum attribute intensity. Each panelist was provided a set of reference foods, water, and crackers per session. Panelists were rewarded with grocery gift cards at the conclusion of each sensory session.

3.2.3. Statistical Analysis

Sensory data was analyzed with analysis of variance (ANOVA) in univariate linear mixed models calculated using the 'lme4' package in Rstudio version 0.99.896 (RStudio, Inc., Boston, MA). Fixed effects were clone, cooking method, and the interaction between clone and cooking method. Random effects were session year, sample order presented to panelists, panelists, and all interactions. Means were estimated using residual maximum likelihood (REML) least square means in attribute mixed models for fixed effects with significant F-values (α=0.05). Clone, cooking method, and clone by cooking method means of significant attributes were contrasted by linear least square mean differences with Welch's adjustments (t-test, α=0.05). Using mixed model clone mean estimates for attributes with significant fixed effects, Spearman's rank correlation coefficients were calculated and clustered by attribute to attribute with z-score normalized Ward's two-way hierarchical clustering analysis (HCA). Clone descriptions using sensory attribute mean estimates were generated in RStudio using value tests (v-tests) through the 'SensoMineR' package, which identifies under or over-represented attributes, or means relatively smaller or greater than the overall mean (modified t-test, α=0.05). Loadings and scores for sensory attributes were Pareto-scaled in principal component analysis (PCA) using SIMCA version 14.1 (MKS Data Analytics Solutions, Umea, Sweden), with component selection guided by Q^2 validation of R^2. Loadings were sensory attributes, whereas scores were raw panelist entries by clone and cooking method. Trends were examined by clone, cooking method, flesh color and year in PCA.

Post-hoc flesh color group means were calculated from mean estimates by clone (e.g. white, yellow, and red or purple) and attributes with significant variation by flesh color group were determined by significant F-values in ANOVA (α=0.05, n=4-6). Purple- and red-fleshed clones were included in the same group to balance flesh color group sizes. Flesh attribute mean differences were compared pairwise by Tukey's Honest Significant Difference (HSD) tests (t-test, α=0.05).

3.3. Results and Discussion

3.3.1. Spearman's Rank Correlations: Positive vs. Negative Attributes

Attributes have weak to strong, negative or positive correlations with one another according to Spearman's rank correlation coefficients. Clustering of correlation coefficients in HCA reveals two clusters of sensory attributes with opposing correlations (Figure 3.1). The larger cluster, cluster 1, encompasses appearance, creamy, fruity, overall quality, sweet, buttery, mealy texture, and potato-like flavor. Cluster 2 includes aroma intensity, earthy, woody, bitter, and off-flavors other than bitter. Considering overall quality as a positive attribute, similarly clustered attributes in cluster 1 are also likely to be positive, whereas attributes in cluster 2 are probably negative. Spearman's rank correlation coefficients highlight proportional variation within positive or negative attribute groups and an inverse relationship between attribute groups.

3.3.2. Linear Mixed Model ANOVA: Attribute Differences by Clone and Cooking Method

Attributes with mean estimate differences by clone are bitter, buttery, creamy, earthy, fruity, off-flavors other than bitter, potato-like flavor, sweet, and woody flavor notes as well as related sensory attributes for appearance, aroma intensity, mealy texture, and overall quality (Table 3.2; Figure 3.2). Comparing positive and negative attributes identified by Spearman's rank correlation coefficients, sensory panelist scores indicate that greater scores for positive attributes and smaller scores for negative attributes are desirable. Sensory attributes with ratings centered on the lower end of the hedonic scale, e.g. off-flavors other than bitter, imply flavor note absence or detection difficulty.

At a glance, purple- and red-fleshed clones have greater ratings for bitter and earthy compared to white- or yellow- fleshed clones. Colored-fleshed clones also have greater appearance scores than white-fleshed clones, which is to be expected since colored-fleshed potatoes are inherently more vibrant and more of a novelty to consumers. Based on Spearman's rank correlation coefficient distinction of positive and negative attributes, Masquerade and Harvest Moon consistently have larger scores for positive attributes and smaller scores for negative attributes compared to other clones.

Attribute differences between cooking methods are minimal. Sensory panelists discerned between cooking methods for creamy and potato-like flavor notes, mealy texture, and overall quality, all of which are rated greater for baked rather than boiled potatoes (Table 3.3; Figure 3.3). Greater creamy, potato-like flavor, and overall quality in baked versus boiled potatoes may be attributed to the slightly longer cooking time required for baking, which enabled more flavor reactions to occur, paired with increased potential of flavor compounds to leach into water during boiling. Furthermore, baking is a drier cooking process than boiling, logically resulting in higher mealy scores. Greater overall quality scores for baked versus boiled potatoes suggests panelists had more positive sensory perceptions of baked rather than boiled potatoes.

Overall quality is the only attribute with significant variation between clone and cooking method interactions for a few clones (Table 3.4). For Russet Burbank, CO4099-3W/Y, and Crimson King, greater overall quality occurred after boiling, suggesting that these clones be boiled to maximize positive sensory perceptions in contrast to CO04063-4R/R, which should be baked to maximize positive perceptions. Yet, clone by cooking method interactions may simply reflect undercooked potatoes in baking sessions versus boiling sessions due to differences in tuber size between clones. Few significant clone by cooking method interactions implies that the influence of cooking method on potato flavor profiles is near negligible.

3.3.3. HCA: Similarities Between Attributes and Clones

Two-way Ward HCA of sensory attributes and clones divides sensory attributes into two main clusters (Figure 3.4). Cluster 1 consists of aroma intensity, appearance, overall quality, and potato-like flavor, all of which are sensory attributes other than flavor notes, with potato-like flavor as an exception. Cluster 2 is mostly flavor notes except for mealy texture, and contains two smaller clusters, off-flavors other bitter, bitter, fruity, and woody flavor notes in cluster 3 and mealy texture, earthy, sweet, buttery, and creamy flavor notes in cluster 4. Expectedly, off-flavors other than bitter clusters closely to bitter in the flavor note group. Another rational cluster is that of sweet, buttery and creamy within cluster 4. Surprisingly, earthy does not cluster closely with woody, despite a sizeable Spearman's rank correlation and intuitively linked sensations.

In the first branch of HCA clustering by clone, advanced selections CO04063-4R/R and Crimson King, both the only red-fleshed clones, separate from the other clones into cluster A. The second branch creates cluster B with Harvest Moon and Masquerade, both of which are yellow-fleshed, half-sib, named cultivars developed by the CSU Potato Breeding and Selection Program. The third branch separates white-fleshed russet clones from colored-fleshed clones, with the exception of Yukon Gold, while remaining branches are mixtures of yellow- and purple-fleshed clones.

For white-fleshed clones, flavor note attribute z-scores (i.e. bitter, buttery, creamy, earthy, fruity, off-flavors other than bitter, potato-like flavor, sweet, woody) appear to be relatively consistent compared to those among yellow-, purple- or red-fleshed potatoes, which is supported by pairwise comparisons in the ANOVA. More consistent ratings for white-fleshed compared to colored-fleshed potatoes is not unexpected since breeding efforts are largely focused on white-fleshed potatoes. Purple- and red-fleshed potatoes especially have received less attention in breeding, where less selection against bitter or other off-flavors caused by phytopigments, glycoalkaloids, and other secondary plant metabolites may be the cause of greater abundances and inconsistencies of negative, off-flavor compounds. Yet, white-fleshed potatoes are not rated lower than yellow-fleshed potatoes for bitter or earthy flavor notes, indicating influences on flavor other than flesh color.

3.3.4. V-Tests: Under- and Over-Represented Attributes per Clone

Although some variation occurs between attribute score mean estimates by clone, clonal variation is most discernible in descriptions generated by v-tests (Table 3.5). In the white flesh group, Russet Norkotah had no prominent descriptors. Russet Burbank, the most common, commercial, white-fleshed russet, has a relatively highly rated potato-like flavor. Russet Nugget has a fairly mealy texture, but no distinctive flavor notes. The recently named cultivar, Fortress Russet, has minimal lemon flavor and high scores for off-flavors other than bitter, possibly related to poor overall quality ratings. Appearance ratings for Fortress Russet were also lowly ranked. The advanced selection, AC00395-2RU, seems promising due to minimal bitter flavors, off-flavors other than bitter, and aroma intensity.

Compared to white-fleshed clone descriptions, yellow-fleshed clones appear to have more diverse flavor notes and a greater buttery flavor versus other flesh colors. Masquerade has low ratings for bitter and off-flavors other than bitter, but high ratings for creamy, fruity and sweet. Harvest Moon also has low scores for bitter flavors and off-flavors other than bitter, with high scores for buttery and low ratings for earthy and woody. Both Masquerade and Harvest Moon have a high overall quality score. Yukon Gold, a common commercial yellow-fleshed potato, does not have prominent sensory attributes other than a low rated earthy flavor, possibly indicating better flavor quality in Masquerade and Harvest Moon. Mealy texture is minimal for Red Luna with no other pronounced attributes. Advanced selection CO04099-3W/Y has no distinguishable attributes, yet advanced selection CO04067-8R/Y has a highly rated buttery flavor and appearance, denoting another promising advanced selection.

Bitter, off-flavors other than bitter, and earthy are most prominent in purple- and red- versus white- or yellow-fleshed clones. By v-test description, Purple Majesty, which is purple-fleshed, is not high in bitter or other off-flavors, but does have a highly rated earthy flavor, appearance, and overall quality. Another purple-fleshed potato clone, advanced selection CO04056-3P/PW, has high scores for bitter, off-flavors other than bitter, earthy, and mealy texture. Red-fleshed advanced selection CO04063-4R/R is high in the same flavor notes plus has low buttery and overall quality scores. Crimson King, also red-fleshed, has high bitter and earthy scores with low buttery, minimally mealy texture, poor appearance and poor overall quality. Although purple- and red-fleshed potatoes may inherently be more bitter and off-flavored due to higher phytopigment content, Crimson King and the advanced selections have a questionable fresh market value since Purple Majesty, a popular commercial cultivar in Colorado, is not characterized with the same negative components (identified through Spearman's rank correlation coefficients) as the advanced selections.

3.3.5. PCA: Positive vs. Negative Attributes in Relation to Clones

The PCA has three components that explain 54.9% of sample variation (R^2), however, a low validation coefficient of 22.2% (Q^2) implies a poor model fit. Still, several sensory attribute loadings have proportional R^2 and Q^2 values, indicating a good model fit for some attributes (Figure 3.5A-D). Scores do not have well-defined separation by clone, cooking method, year, order, or flesh color, yet, due to the subjective nature of flavor, some separation occurs between panelists, which is an important reminder of personal preferences and the subjective nature of flavor. Sensory attribute loadings correlate in two distinct clusters in a normalized correlation biplot of PC2 versus PC1 (Figure 3.5E). The top left cluster, cluster 1, contains overall quality, sweet, buttery, creamy, potato-like flavor, with weak correlations to fruity and appearance, whereas cluster 2 contains off-flavors other than bitter, lemon, umami, aroma intensity, woody, earthy, and bitter with a weak correlation to mealy texture. Using the same logic for identifying positive and negative attributes in Spearman's rank coefficient correlation HCA, cluster 1, which is closely associated with PC1, contains positive attributes whereas cluster 2, related to PC2, contains negative attributes. Though there are correlations within clusters, there is no correlation between clusters. Several scores for CO04056-3P/PW, CO04067-8R/Y, CO4099-3W/Y, Harvest Moon, and Yukon Gold correlate with the positive attribute cluster, while multiple scores for CO04056-3P/PW, CO04063-4R/R, CO4099-3W/Y, Crimson King, Fortress Russet, Purple Majesty, and Russet Nugget correlate with the negative attribute cluster.

3.3.6. Post-Hoc ANOVA: Attribute Differences by Flesh Color

In flesh color group comparisons, bitter, buttery, earthy, off-flavors other than bitter, woody, appearance, and aroma intensity attributes have mean score differences (Table 3.6; Figure 3.6), implying that flesh color is somewhat indicative of flavor profiles. Purple- and red-fleshed potatoes have greater ratings for bitter, earthy, and aroma intensity relative to white- or yellow- fleshed potatoes, as well as greater ratings for off-flavors other than bitter and woody compared to yellow-fleshed potatoes. Buttery flavor scores are greater for yellow-fleshed potatoes versus other flesh colors. Additionally, appearance is greater for yellow- versus white-fleshed potatoes.

3.3.7. Summary of Positive vs. Negative Attributes

Positive attributes consistent across Spearman's rank correlation coefficient HCA and PCA are appearance, buttery, creamy, fruity, sweet, overall quality, and potato-like flavor. Consistent negative attributes are aroma intensity, bitter, earthy, off-flavors other than bitter, and woody. Mealy texture was a positive attribute in Spearman correlation coefficient HCA but a negative attribute in PCA. The HCA for sensory attributes and clone mean estimates did not follow a pattern similar to Spearman's rank correlation coefficient HCA and PCA.

3.3.8. Application of Results

Flavor profiles generated from sensory analysis are informative to producers that are interested in growing fresh market potatoes with high flavor quality, which should increase profit since more flavorful produce tends to have a higher value than produce with poor flavor quality.[9] Flavor profiles may also be useful to consumers that wish to maximize flavor quality or select a product with specific flavor components. For selection-based breeding programs, such as the Colorado State University Potato Breeding and Selection Program, flavor profiles may be used to guide parental crosses for crop flavor quality improvement. This sensory analysis can also guide flavor compound biomarker discovery and development of efficient biomarker assays, therefore enabling selection for phenotypes with improved flavor. Flavor biomarker identification will further provide a framework for potato flavor gene studies.

3.4. Summary

The purpose of this study was to examine fresh market potato flavor diversity and to identify relevant sensory attributes. A sample population of 15 clones with various skin and flesh

colors was described by and scored for 15 attributes using a trained panel. Clones had significant differences for 13 of 15 attributes according to linear mixed model ANOVA (F-test, α=0.05) and least square mean differences (t-test, α=0.05). The influence of cooking method on flavor was minimal. Positive attributes identified in Spearman's correlation coefficient hierarchical clustering analysis and principle component analysis were appearance, buttery, creamy, fruity, sweet, potato-like flavor, and overall quality, whereas negative attributes were aroma intensity, bitter, earthy, off-flavors other than bitter, and woody. The most positively perceived clones across statistical methods were Masquerade and Harvest Moon, which are yellow-fleshed half sibs developed by the Colorado Potato Breeding and Selection Program.

Table 3.1. Reference foods selected by a trained sensory panel for cooked potato flavor description with a hedonic scale (1 to 9, absence to maximum intensity)

Flavor	**Reference Food**
Sweetness	Sugar cube
Fruity Flavor	Apple slice
Lemon Flavor	Lemon slice
Umami Flavor	Chicken broth
Buttery Flavor	Butter slice
Creamy Flavor	White chocolate
Earthy Flavor	Beet slice
Woody Flavor	Toothpick
Bitter	Tonic water

Table 3.2. Hedonic score (1 to 9, absence to maximum intensity) clone mean estimates[a] and comparisons[b] of cooked potato sensory attributes rated by a trained sensory panel

Flesh Color	Clone	Bitter			Buttery			Creamy			Earthy			Fruity		
White	AC00395-2RU	1.53	[0.84, 2.23]	A	2.66	[1.83, 3.48]	BCD	2.51	[1.70, 3.31]	AB	3.09	[2.29, 3.88]	ABC	1.97	[1.34, 2.60]	ABCD
	Fortress Russet	2.21	[1.52, 2.91]	BCD	2.79	[1.96, 3.61]	CD	2.87	[2.07, 3.68]	BCDE	3.53	[2.74, 4.32]	BCD	1.84	[1.20, 2.47]	A
	Russet Burbank	2.39	[1.68, 3.10]	CDE	2.41	[1.58, 3.24]	ABC	2.26	[1.45, 3.08]	A	3.47	[2.67, 4.27]	ABCD	2.07	[1.43, 2.71]	ABCD
	Russet Norkotah	1.71	[1.02, 2.41]	AB	2.79	[1.96, 3.61]	CD	2.53	[1.72, 3.34]	AB	3.64	[2.85, 4.43]	CDE	1.89	[1.26, 2.52]	AB
	Russet Nugget	1.89	[1.18, 2.59]	ABC	2.54	[1.71, 3.37]	BC	2.54	[1.73, 3.35]	AB	3.23	[2.44, 4.03]	ABCD	2.02	[1.38, 2.65]	ABCD
Yellow	CO04067-8R/Y	1.91	[1.21, 2.62]	ABC	3.33	[2.50, 4.16]	E	3.12	[2.30, 3.93]	CDE	3.47	[2.68, 4.27]	ABCD	2.03	[1.39, 2.67]	ABCD
	CO04099-3W/Y	2.41	[1.70, 3.11]	CDE	2.66	[1.83, 3.49]	BCD	2.43	[1.62, 3.24]	AB	3.57	[2.78, 4.37]	BCD	2.28	[1.64, 2.92]	BCDE
	Masquerade	1.58	[0.89, 2.28]	A	3.14	[2.31, 3.96]	DE	3.32	[2.51, 4.13]	E	3.12	[2.33, 3.91]	ABCD	2.63	[2.00, 3.27]	E
	Harvest Moon	1.68	[0.98, 2.38]	AB	3.35	[2.53, 4.18]	E	3.24	[2.43, 4.05]	DE	2.96	[2.16, 3.75]	A	1.90	[1.27, 2.54]	ABC
	Red Luna	2.47	[1.77, 3.17]	CDE	2.76	[1.93, 3.58]	CD	2.86	[2.05, 3.67]	BCDE	3.58	[2.79, 4.37]	BCD	2.47	[1.83, 3.11]	DE
	Yukon Gold	1.92	[1.23, 2.61]	ABC	2.89	[2.06, 3.71]	CDE	2.53	[1.73, 3.34]	AB	3.03	[2.25, 3.82]	AB	2.43	[1.79, 3.06]	DE
Purple	CO04056-3P/PW	2.85	[2.14, 3.56]	E	2.53	[1.70, 3.36]	ABC	2.55	[1.73, 3.36]	AB	4.39	[3.59, 5.19]	F	2.23	[1.59, 2.87]	ns
	Purple Majesty	2.10	[1.40, 2.80]	ABC	2.76	[1.93, 3.58]	CD	2.75	[1.94, 3.56]	ABCD	4.17	[3.38, 4.97]	EF	2.17	[1.54, 2.81]	ABCD
Red	CO04063-4R/R	3.83	[3.02, 4.64]	F	1.91	[1.01, 2.81]	A	2.56	[1.66, 3.46]	ABC	4.44	[3.55, 5.34]	F	1.97	[1.25, 2.68]	ABCD
	Crimson King	2.82	[2.11, 3.53]	DE	2.16	[1.33, 2.99]	AB	2.60	[1.79, 3.42]	ABC	3.68	[2.88, 4.47]	DE	2.35	[1.71, 2.99]	CDE

Flesh Color	Clone	Off Flavors Other Than Bitter			Potato-Like			Sweet			Woody		
White	AC00395-2RU	1.19	[0.79, 1.58]	A	5.99	[5.48, 6.50]	CDE	2.78	[2.06, 3.50]	BC	2.48	[1.78, 3.19]	BCDEF
	Fortress Russet	2.00	[1.61, 2.40]	CD	5.62	[5.11, 6.13]	BCD	2.20	[1.48, 2.93]	A	2.72	[2.02, 3.42]	EF
	Russet Burbank	1.54	[1.14, 1.95]	AB	6.28	[5.76, 6.80]	E	2.37	[1.65, 3.10]	AB	2.51	[1.80, 3.22]	BCDEF
	Russet Norkotah	1.53	[1.14, 1.92]	AB	5.88	[5.38, 6.39]	CDE	2.41	[1.69, 3.14]	AB	2.54	[1.83, 3.24]	CDEF
	Russet Nugget	1.33	[0.93, 1.74]	A	5.99	[5.48, 6.51]	CDE	2.28	[1.55, 3.00]	AB	2.17	[1.46, 2.88]	ABCD
Yellow	CO04067-8R/Y	1.43	[1.03, 1.84]	AB	6.00	[5.48, 6.52]	CDE	2.54	[1.81, 3.27]	ABC	2.40	[1.69, 3.11]	BCDE
	CO04099-3W/Y	1.56	[1.16, 1.96]	ABC	5.58	[5.06, 6.10]	ABCD	2.55	[1.82, 3.28]	ABC	2.63	[1.92, 3.34]	DEF
	Masquerade	1.20	[0.80, 1.59]	A	5.81	[5.29, 6.32]	BCDE	3.40	[2.67, 4.12]	D	2.03	[1.33, 2.74]	AB
	Harvest Moon	1.17	[0.77, 1.57]	A	5.97	[5.46, 6.48]	CDE	2.64	[1.91, 3.36]	ABC	1.90	[1.19, 2.06]	A
	Red Luna	1.47	[1.07, 1.86]	AB	5.45	[4.94, 5.97]	ABC	2.64	[1.91, 3.36]	ABC	2.36	[1.65, 3.06]	ABCDE
	Yukon Gold	1.50	[1.10, 1.89]	AB	5.74	[5.24, 6.25]	BCD	2.97	[2.25, 3.69]	C	2.06	[1.36, 2.77]	ABC
Purple	CO04056-3P/PW	1.89	[1.48, 2.30]	BC	5.34	[4.82, 5.86]	AB	2.23	[1.50, 2.95]	AB	2.92	[2.21, 3.63]	F
	Purple Majesty	1.52	[1.12, 1.92]	AB	6.08	[5.56, 6.59]	DE	2.67	[1.94, 3.39]	ABC	2.74	[2.03, 3.44]	EF
Red	CO04063-4R/R	2.46	[1.94, 2.99]	D	5.38	[4.72, 6.03]	ABC	2.21	[1.41, 3.01]	AB	2.52	[1.73, 3.31]	BCDEF
	Crimson King	2.00	[1.60, 2.41]	CD	5.09	[4.57, 5.61]	A	2.28	[1.55, 3.01]	AB	2.60	[1.89, 3.31]	DEF

Flesh Color	Clone	Appearance			Aroma Intensity			Mealy Texture			Overall Quality		
White	AC00395-2RU	5.75	[5.13, 6.37]	BCDE	4.26	[3.40, 5.12]	A	3.88	[3.06, 4.69]	EF	6.04	[5.56, 6.51]	CDEF
	Fortress Russet	4.90	[4.28, 5.52]	A	4.84	[3.98, 5.70]	CD	3.22	[2.41, 4.03]	ABCD	5.34	[4.87, 5.81]	AB
	Russet Burbank	5.48	[4.85, 6.12]	ABC	4.97	[4.10, 5.84]	CD	3.30	[2.48, 4.12]	ABCDE	5.46	[4.98, 5.94]	BC
	Russet Norkotah	5.65	[5.02, 6.27]	BCD	4.74	[3.87, 5.60]	ABC	3.46	[2.64, 4.27]	BCDE	5.55	[5.08, 6.02]	BC
	Russet Nugget	5.50	[4.86, 6.13]	ABC	4.79	[3.92, 5.65]	ns	4.66	[3.84, 5.48]	G	5.89	[5.41, 6.37]	CDE
Yellow	CO04067-8R/Y	6.43	[5.80, 7.07]	F	4.67	[3.80, 5.54]	ABC	3.90	[3.08, 4.73]	EF	6.14	[5.65, 6.62]	DEF
	CO04099-3W/Y	6.27	[5.64, 6.90]	DEF	4.83	[3.96, 5.70]	BCD	3.58	[2.76, 4.40]	CDEF	5.63	[5.15, 6.11]	BC
	Masquerade	6.22	[5.60, 6.85]	DEF	4.95	[4.08, 5.81]	CD	3.62	[2.81, 4.44]	DEF	6.48	[6.01, 6.96]	F
	Harvest Moon	6.26	[5.64, 6.89]	DEF	4.30	[3.43, 5.16]	AB	2.98	[2.16, 3.79]	ABC	6.14	[5.66, 6.61]	DEF
	Red Luna	6.27	[5.65, 6.90]	EF	4.81	[3.95, 5.68]	BCD	2.87	[2.05, 3.68]	AB	5.53	[5.06, 6.01]	BC
	Yukon Gold	5.94	[5.32, 6.56]	CDEF	4.64	[3.78, 5.50]	ABC	3.73	[2.92, 4.54]	DEF	5.66	[5.19, 6.14]	BCD
Purple	CO04056-3P/PW	6.19	[5.55, 6.82]	DEF	5.30	[4.43, 6.17]	D	4.10	[3.28, 4.93]	FG	5.84	[5.35, 6.32]	CDE
	Purple Majesty	6.25	[5.63, 6.88]	DEF	5.16	[4.30, 6.03]	CD	3.81	[3.00, 4.63]	DEF	6.33	[5.85, 6.80]	EF
Red	CO04063-4R/R	5.92	[5.18, 6.65]	BCDEF	5.27	[4.32, 6.22]	CD	3.39	[2.46, 4.32]	ABCDEF	4.80	[4.21, 5.38]	A
	Crimson King	5.24	[4.60, 5.87]	AB	5.10	[4.23, 5.97]	CD	2.82	[2.00, 3.64]	A	4.85	[4.37, 5.34]	A

[a]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=17-38 hedonic scores x 15 clones).

[b]All sensory attribute clone means are significantly different according to mixed model ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Welch's adjusted t-tests (α=0.05).

Table 3.3. Hedonic score (1 to 9, absence to maximum intensity) cooking method mean estimates with 95% confidence intervals[a] and comparisons[b] of cooked potato sensory attributes rated by a trained sensory panel

	Baked		Boiled	
Creamy	2.99	[2.24, 3.74]	2.43	[1.67, 3.19]
Potato-Like	6.06	[5.66, 6.45]	5.44	[5.03, 5.85]
Mealy Texture	4.01	[3.27, 4.75]	3.10	[2.34, 3.87]
Overall Quality	5.95	[5.55, 6.35]	5.47	[5.04, 5.90]

[a]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=237-291 x 2 cooking methods).

[b]Cooking method means by sensory attribute are significantly different according to mixed model ANOVA F-tests (α=0.05) and pairwise comparison of means in Welch's adjusted t-tests (α=0.05).

Table 3.4. Hedonic score (1 to 9, absence to maximum intensity) clone by cooking method mean estimates with 95% confidence intervals[a] and comparisons[b] of cooked potato overall quality rated by a trained sensory panel

Flesh Color	Clone	Baked		Boiled	
White	**Russet Burbank**	4.45	[3.81, 5.09]	6.14	[5.54, 6.73]
Yellow	**CO04099-3W/Y**	5.67	[4.92, 6.42]	6.27	[5.63, 6.90]
Red	**CO04063-4R/R**	6.01	[5.41, 6.61]	5.53	[4.91, 6.15]
	Crimson King	3.87	[3.07, 4.66]	5.67	[5.08, 6.25]

[a]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=8-21 x 2 cooking methods x 15 clones).

[b]Cooking method means by clone for overall quality are significantly different according to a mixed model ANOVA F-test (α=0.05) and pairwise comparison of means for some clones in Welch's adjusted t-tests (α=0.05).

Table 3.5. Cooked potato clone description using sensory attributes identified by v-tests[a] with hedonic score (1 to 9, absence to maximum intensity) mean estimates[b] calculated from ratings of a trained sensory panel

Flesh Color	Clone	Flavor Notes	Other
White	**AC00395-2RU**	Low bitterness, low off-flavors other than bitter	Low aroma intensity
	Fortress Russet	Low lemon flavor, high off-flavors other than bitter	Poor appearance, poor overall quality
	Russet Norkotah	None	None
	Russet Burbank	High potato-like flavor	None
	Russet Nugget	None	Highly mealy texture
Yellow	**CO04067-8R/Y**	High butteriness	Great appearance
	CO04099-3W/Y	None	None
	Harvest Moon	Low bitterness, high butteriness, low earthiness, low off-flavors other than bitter, low woodiness	Highly mealy texture, high overall quality
	Masquerade	Low bitterness, high creaminess, high frutiness, high sweetness, low off-flavors other than bitter	High overall quality
	Red Luna	None	Minimally mealy texture
	Yukon Gold	Low earthiness	
Purple	**CO04056-3P/PW**	High bitterness, high earthiness, high off-flavors other than bitter	Highly mealy texture
	Purple Majesty	High earthiness	Great appearance, high overall quality
Red	**CO04063-4R/R**	High bitterness, low butteriness, high earthiness, high off-flavors other than bitter	Poor overall quality
	Crimson King	High bitterness, low butteriness, high earthiness	Poor appearance, minimally mealy texture, poor overall quality

[a]A v-test is a modified t-test used for the identification of over or under-represented variables, or in this case, clone means significantly smaller or greater (α=0.05) than the overall mean of a sensory attribute

[b]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=17-38 hedonic scores x 15 clones).

Table 3.6. Hedonic score (1 to 9, absence to maximum intensity) flesh color mean estimates with 95% confidence intervals[a] and comparisons[b] of cooked potato sensory attributes rated by a trained sensory panel

		Flavor Notes														
Flesh Color	**n**	**Bitter**			**Buttery**			**Earthy**			**Off-Flavors Other Than Bitter**			**Woody**		
White	5	1.95	[1.64, 2.26]	A	2.64	[2.50, 2.78]	AB	3.39	[3.19, 3.59]	A	1.52	[1.25, 1.79]	AB	2.48	[2.31, 2.66]	AB
Yellow	6	2.00	[1.70, 2.29]	A	3.02	[2.78, 3.26]	B	3.29	[3.06, 3.52]	A	1.39	[1.26, 1.52]	A	2.23	[2.01, 2.45]	A
Purple and Red	4	2.90	[2.21, 3.60]	B	2.34	[1.97, 2.71]	A	4.17	[3.83, 4.51]	B	1.97	[1.59, 2.35]	B	2.70	[2.53, 2.86]	B

		Other Characteristics					
Flesh Color	**n**	**Appearance**			**Aroma Intensity**		
White	5	5.45	[5.17, 5.74]	A	4.72	[4.48, 4.95]	A
Yellow	6	6.23	[6.10, 6.36]	B	4.70	[4.52, 4.88]	A
Purple and Red	4	5.90	[5.44, 6.35]	AB	5.21	[5.12, 5.30]	B

[a]Flesh group mean of REML mean estimates by clone (n=5 for white, n=6 for yellow, n=4 for red) from a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects.

[b]Flesh group means by sensory attribute are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05).

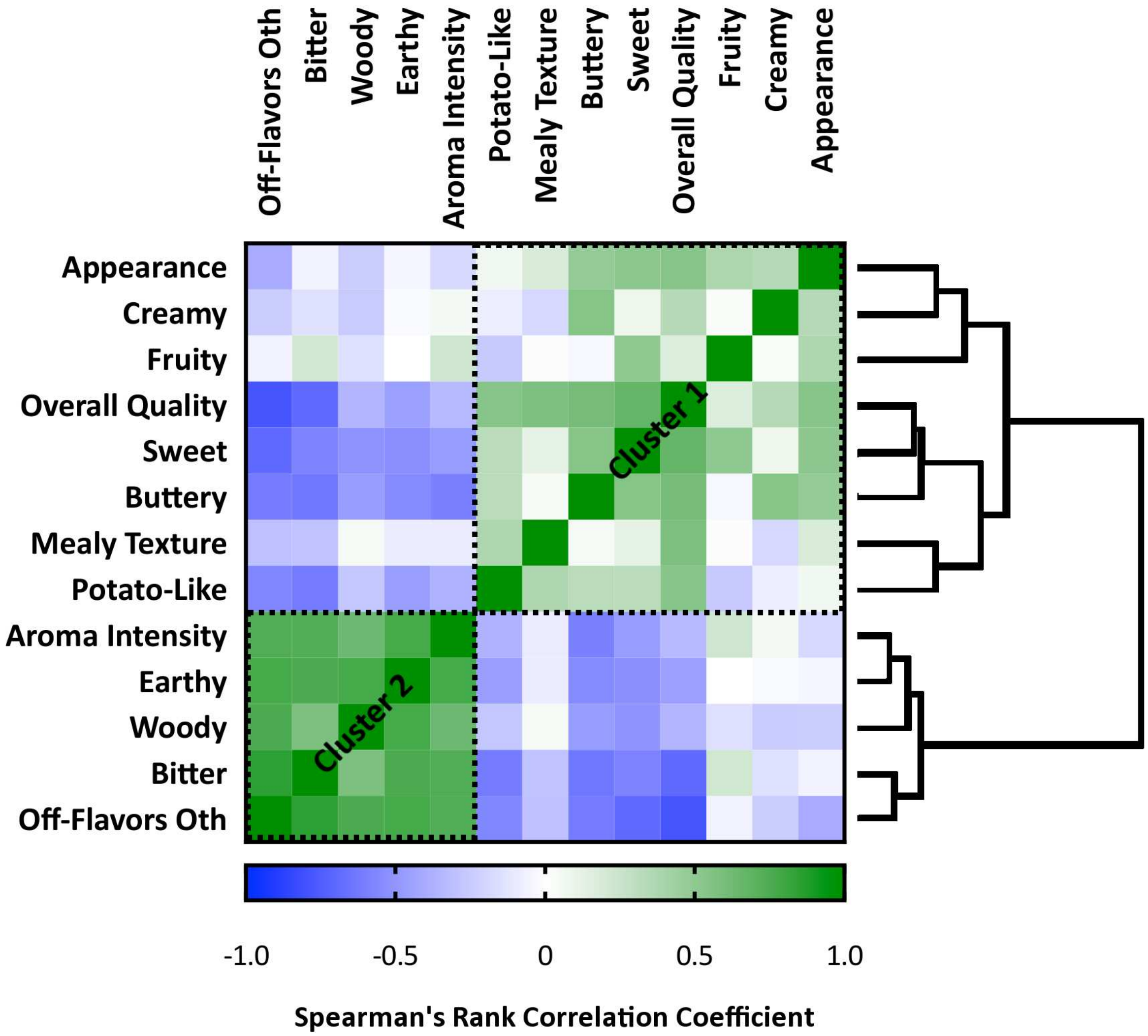

Figure 3.1. Spearman's rank correlation coefficients with two-way Ward HCA of cooked potato sensory attributes using clone hedonic score (1 to 9, absence to maximum intensity) mean estimates[a] calculated from ratings of a trained sensory panel

[a]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=17-38 hedonic scores x 15 clones).

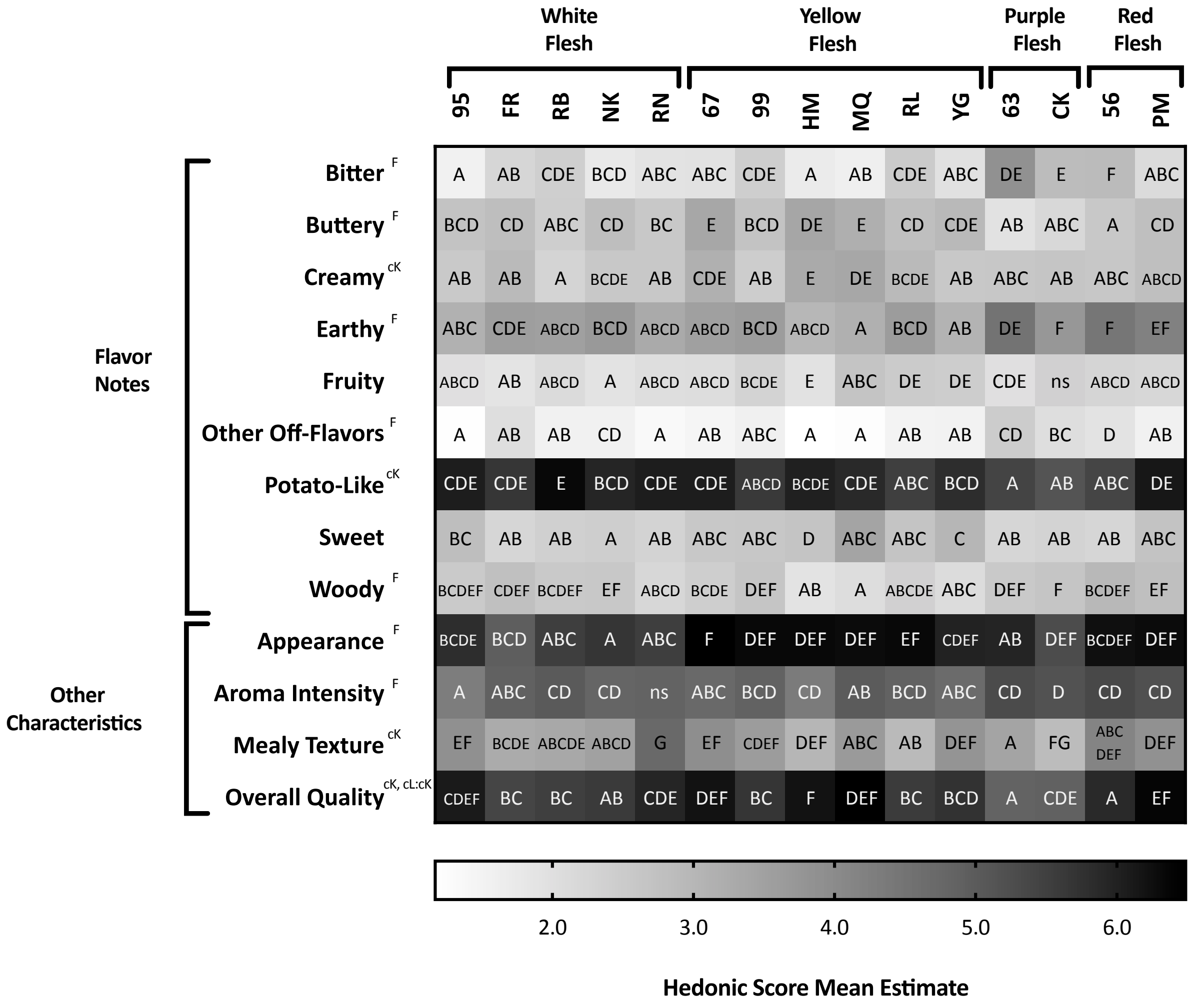

Figure 3.2. Hedonic score (1 to 9, absence to maximum intensity) clone[a] mean estimates[b] and comparisons[c] of cooked potato sensory attributes rated by a trained sensory panel

[a]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, 63=CO04063-4R/R, CK=Crimson King.

[b]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=17-38 hedonic scores x 15 clones).

[c]All sensory attribute clone means are significantly different according to mixed model ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Welch's adjusted t-tests (α=0.05). Sensory attributes are also significantly different according to mixed model F-tests (α=0.05) for cK=cooking method, cL:cK=clone by cooking method, or F=flesh color when notated.

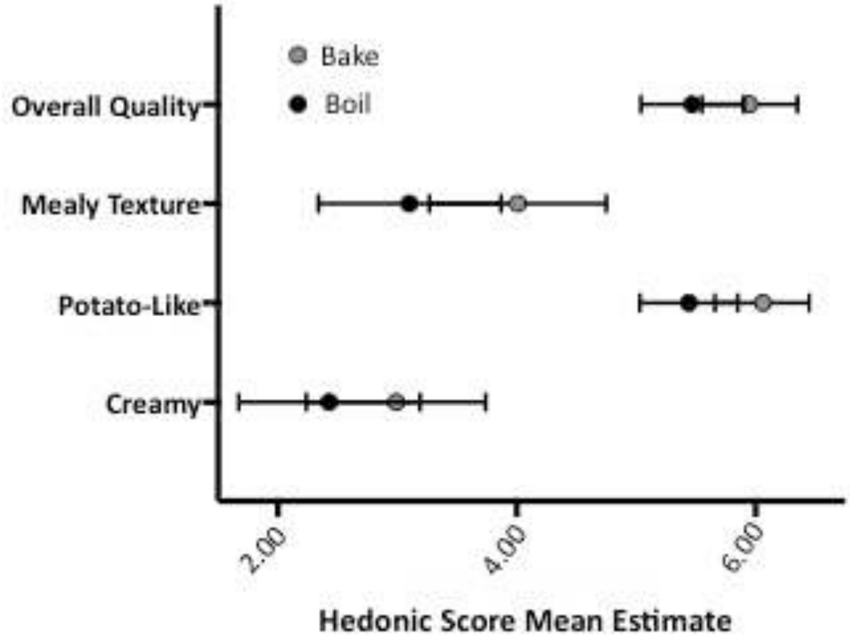

Figure 3.3. Hedonic score (1 to 9, absence to maximum intensity) cooking method mean estimates with 95% confidence intervals[a] and comparisons[b] of cooked potato sensory attributes rated by a trained sensory panel

[a]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=237-291 hedonic scores x 2 cooking methods).

[b]Cooking method means by sensory attribute are significantly different according to mixed model ANOVA F-tests (α=0.05) and pairwise comparison of means in Welch's adjusted t-tests (α=0.05).

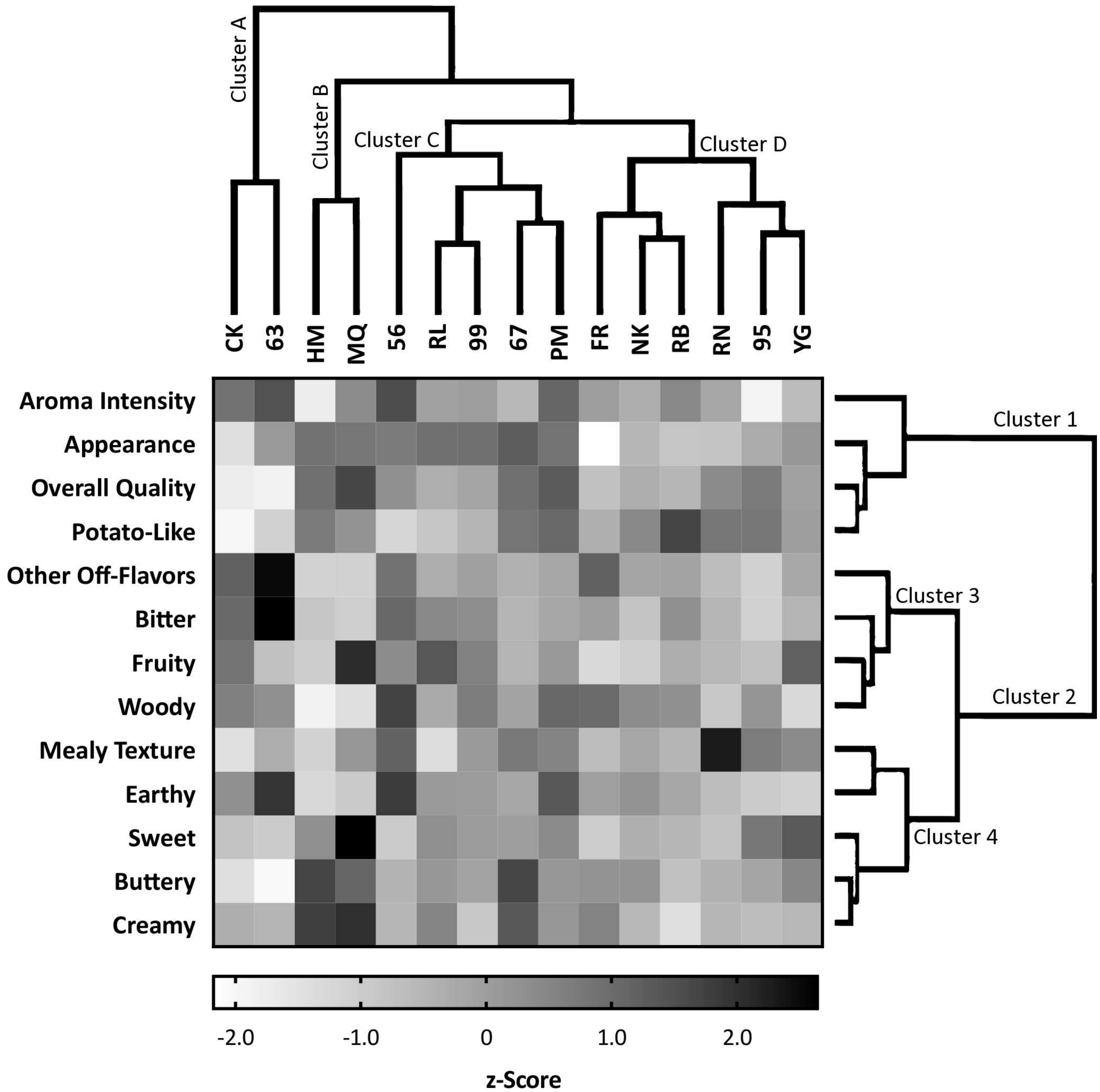

Figure 3.4. Hedonic score (1 to 9, absence to maximum intensity) clone[a] mean estimates[b] with z-score normalization and two-way Ward HCA of cooked potato sensory attributes[c] rated by a trained sensory panel

[a]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, 63=CO04063-4R/R, CK=Crimson King.

[b]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=17-38 hedonic scores x 15 clones).

[c]All sensory attribute clone means are significantly different according to mixed model ANOVA F-tests (α=0.05).

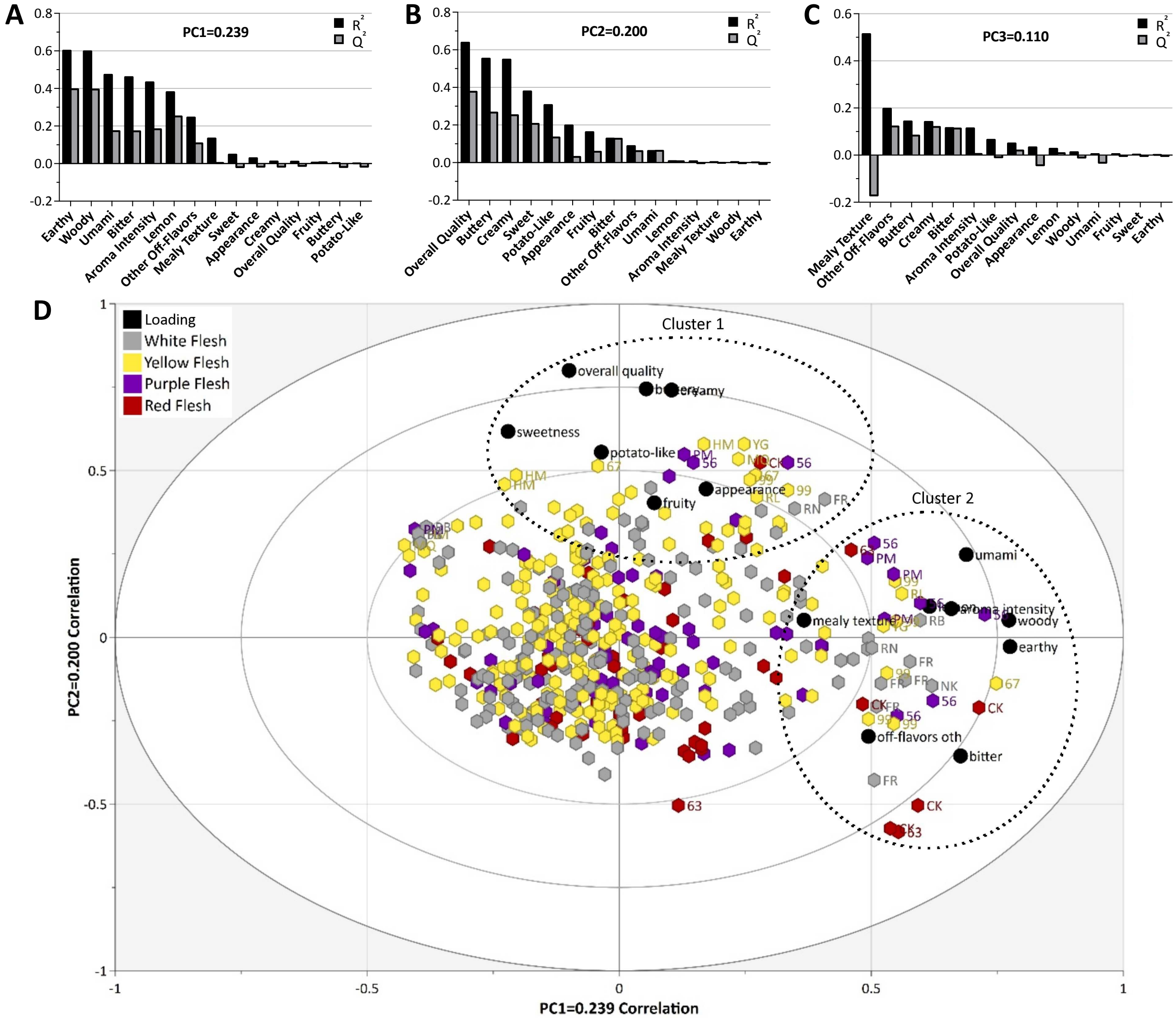

Figure 3.5. (A-C) Loading contributions and (D) normalized correlation R^2 biplot in principle component analysis of Pareto-scaled sensory attribute[a] loadings and clone[b] by cooking method hedonic scores colored by clone flesh color[c]

[a]All sensory attributes are significantly different according to mixed model ANOVA F-tests (α=0.05) for clone, cooking method, or clone and cooking method interactions (n=8-21 hedonic scores x 2 cooking methods x 15 clones).

[b]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, 63=CO04063-4R/R, CK=Crimson King.

[c]Only scores with $R^2 \geq 0.50$ are labeled with clone codes

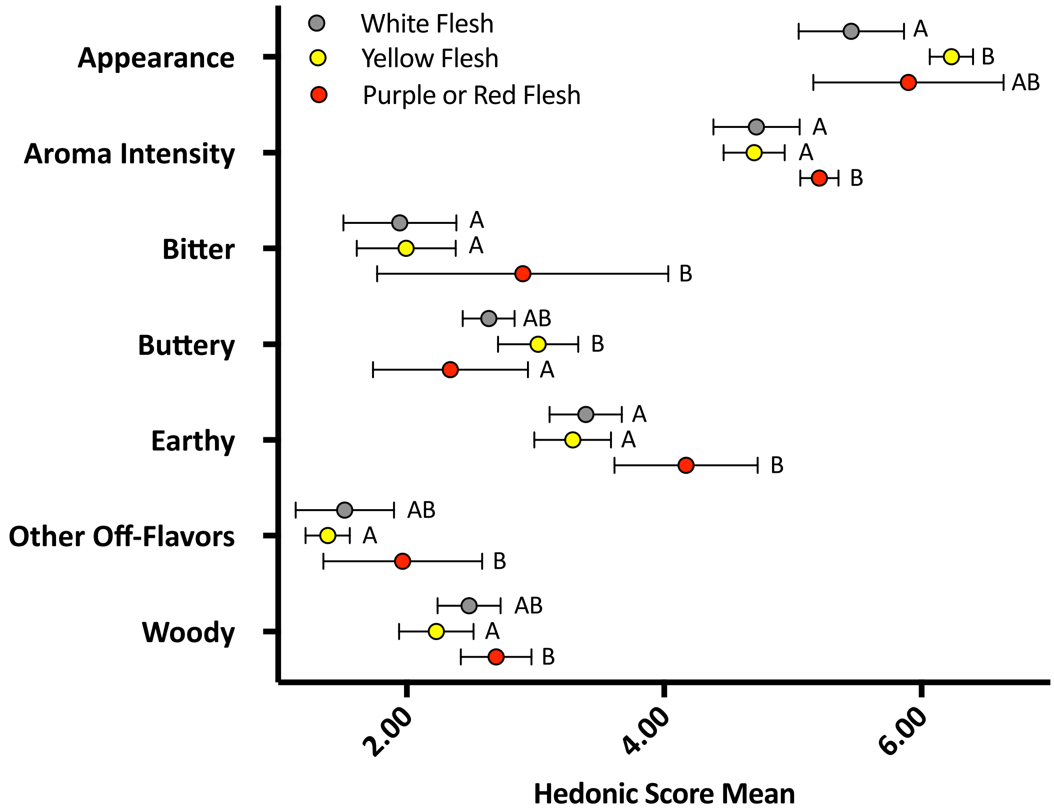

Figure 3.6. Hedonic score (1 to 9, absence to maximum intensity) flesh color mean estimates with 95% confidence intervals[a] and comparisons[b] of cooked potato sensory attributes rated by a trained sensory panel

[a]Flesh group mean of REML mean estimates by clone (n=5 for white, n=6 for yellow, n=4 for red) from a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects.

[b]Flesh group means by sensory attribute are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05).

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CHAPTER 4. NON-TARGETED AROMA COMPOUND ANALYSIS

4.1. Introduction

Flavor is a subjective, complex quality trait that primarily involves taste and aroma sensations, although appearance, texture, or other characteristics may influence perception.[1] Taste is the basic reception of bitter, sweet, salty, sour, and umami on the tongue.[2] In potatoes, umami and bitter are most prominent, whereas the other tastes are minimal.[3] Aroma, detected in the olfactory, includes more diverse sensations that typically contribute to flavor to a greater extent than taste,[1] particularly for potatoes.[3] Aroma flavor metabolites and products range from volatile to semi-volatile, thus are potent even in small concentrations.[4]

Since the late 1960s, dozens of aroma flavor metabolites and products have been identified in potatoes. Most potato aroma compounds result from interactions during cooking between fatty acid, sugar, and amino acid precursors, including Strecker amino acid degradation to form aldehydes, lipid degradation to form aldehydes and ketones, Maillard reactions that form pyrazines, and other sugar degradation reactions.[5,6] Other aroma metabolites include sulfur compounds[6,7] as well as terpenes and pyrazines present in raw tubers.[10] Although overall flavor is synergistic between metabolites and products,[4] pyrazines[8-10] and aldehydes[6,11] may be particularly influential to potato flavor. Aroma compounds differ by cultivar,[12-14] environment,[13] storage conditions,[6,11] and cooking method.[14]

Compared to older volatile extraction methods, headspace solid-phase microextraction (HS-SPME) has similar sensitivity, greater automation capacity, and faster extraction times.[15] Solvents are also not used, reducing chemical waste and error likelihood. This method has been used in a handful of European fresh market potato volatile metabolite studies coupled to gas chromatography and mass spectrometry (GC-MS),[15-19] however, has not been applied to American fresh market potato cultivars or a sample group with differential cooking methods. Assessment of potato aroma metabolites and products using HS-SPME will enable tentative identification of American potato flavor compounds and serve as an indicator for method application to high-throughput flavor phenotyping. Confirmation of potato flavor aroma compounds with sensory analysis may elucidate target flavor compounds for selection-based breeding programs. In the following study, aroma compounds were extracted through HS-SPME from American potatoes for evaluation with GC-MS across fifteen clones and two cooking methods.

4.2. Materials and Methods

4.2.1. Plant Material

Potato clones were field grown at the Colorado State University San Luis Valley Research Center (SLVRC, Center, CO) using conventional production practices in 2014 and 2015. Tuber material consisted of fifteen clones, including five advanced breeding selections and ten named cultivars. White-fleshed clones were AC00395-2RU, Fortress Russet, Russet Burbank, Russet Norkotah, and Russet Nugget; yellow-fleshed clones were CO04067-8R/Y, CO04099-3W/Y, Harvest Moon, Masquerade, Red Luna, and Yukon Gold; purple-fleshed clones were CO04056-3P/PW and Purple Majesty; red-fleshed clones were CO04063-4R/R and Crimson King. In the beginning of the year following each growing season, tubers were removed from storage and baked or boiled. Cooked potatoes were diced, subsequently frozen with liquid N_2, and stored at -80°C until volatile analysis. Bulked, randomized samples for each clone by cooking method contained material from at least three tubers.

4.2.2. Aroma Compound Reference Standards

Although samples were analyzed using a non-targeted metabolomics workflow, some known potato flavor analytes were diluted to 30 ng μL^{-1} with 100% methanol as a standard mix to facilitate method development for gas chromatography coupled to mass spectrometry (GC-MS) and confirm identifications when appropriate. Exceptions were α-copaene and dimethyl trisulfide, diluted to 14 and 15 ng μL^{-1}, respectively. All standards were produced by the Sigma-Aldrich Company, LLC (St. Louis, MO): 3-carene (90%), α-copaene (≥90%), β-damascenone (≥90%), 1-decanal (≥98%), hexadecane (≥99.8%), furfural (≥98%), isovaleraldehyde (≥97%), α-limone (≥99%), methional (≥97%), 1-octen-3-ol (98%), 2-pentanone (99.5%), 2-phenylacetylaldehyde (≥90%), α-pinene (99%+), 2,5-dimethylpyrazine (98%), 2-isobutyl-3-methoxypyrazine (99%), 2-isopropyl-3-methoxypyrazine (97%), dimethyl disulfide (≥99%), and dimethyl trisulfide (≥98%).

4.2.3. HS-SPME GC-MS

In both years, 5.0 g cooked potato samples were transferred to 20 mL (75.5 x 22.5 mm; MicroSolv Technology Co., Leland, NC) HS vials. Five randomized technical replications per clone and cooking method were gently thawed to room temperature, mashed within HS vials, resealed, and placed into an 80°C water bath for 5 minutes to equilibrate vial and HS temperatures. A 75 µm carboxen-coated 23 mm polydimethylsiloxane fiber (CAR/PDMS, Supelco Inc., Bellefonte, PA) was used for SPME of volatile compounds. Adsorption occurred for 20 minutes at 50 °C and 300 rpm, followed by desorption and splitless injection of metabolites and products with helium carrier gas at a constant flow rate of 1.5 mL min^{-1} for 2 minutes at 280°C. An Agilent 7890A (Agilent Technologies, Santa Clara, CA) equipped with an Agilent VF5-ms GC column separated analytes using temperature ramps from 35 to 280°C. An

Agilent 240 ion trap detected analyte fragments after internal electron ionization at 70 eV and a full scan mode from 29-400 m/z at 0.45 seconds per scan. Automation was achieved through System Control in MS Workstation version 6 (Agilent Technologies).

4.2.4. Data Processing

Raw GC/MS chromatographs were processed with the XCMS package in RStudio version 0.99.896 (RStudio, Inc., Boston, MA) using a CSU Proteomics and Metabolomics Facility chromatography processing workflow with supporting functions. Processing included peak detection, grouping, retention time correction, filling, and normalization using the total ion current (TIC). A signal to noise ratio (S/N) of 2 was used for peak detection. Using the 'ramclustR' package in RStudio, peak data was clustered and correlated by trends in peak abundance and retention time to generate reconstructed spectra for tentative identification and statistical analysis. Reconstructed peak areas were analyzed in an analysis of variance (ANOVA) with false discovery rate (FDR) adjustments (α=0.05) and principal component analysis (PCA) with Pareto-scaling (α=0.05). Mass spectra of compound peaks with significant differences in ANOVA and PCA were tentatively identified using minimum intensity matching of 450 in the National Institute of Standards and Technology (NIST) MS Search Software version 2.0f (NIST, Gaithersburg, MD) with the NIST 11 database. A user-generated database of standards confirmed some tentative spectral identifications with retention time matching.

4.2.5. Statistical Analysis

Compound peak areas were log-transformed prior to statistical analyses. Spearman's rank correlation coefficients between compounds across both years were calculated and clustered by Ward's two-way hierarchical clustering analysis (HCA). Two-way ANOVA was conducted for each identified compound by clone, cooking method, and their interaction. For compounds with a significant F-value in ANOVA (α=0.05), means were compared pairwise by clone and the interaction of clone and cooking method using Tukey's Honest Significant Difference (HSD) tests (t-test, α=0.05) or pairwise with Welch's adjusted t-tests (α=0.05) for cooking method comparison. Due to differences in compounds detected, two-way ANOVA was performed by individual sample year rather than pooled between years. Compounds with significant variation in two-way ANOVA were subject to z-score normalization for Ward's two-way HCA by clone. The SIMCA version 14.1 program (MKS Data Analytics Solutions, Umea, Sweden) was used to conduct Pareto-scaled multivariate PCA with clone by cooking method compound peak areas as variables for individual and collective sample years. Components were selected using Q^2 validation of R^2.

Clone peak means were combined by respective flesh color into the following groups: white, yellow, and purple or red flesh. Purple- and red-fleshed clones were included in the same group to balance flesh color group sizes. A post-hoc analyte comparison between flesh colors was evaluated with ANOVA and Tukey's HSD tests (α=0.05, n=4-6).

4.3. Results and Discussion

4.3.1. Data Processing: Total Peaks to Identifiable Peaks with Variation Across Samples

Processing produced 539 analyte peaks in 2014 and 583 in 2015. In the 2014 ANOVA with FDR adjustments, 3.90%, 1.67%, and 0.19% of analytes had significant p-values (α=0.05) by clone, cooking method, and their interaction, respectively. Percentages of analytes with significant p-values for analogous ANOVA treatments in 2015 were 77.74%, 30.98%, and 38.40%. Across 20 principal components in PCA, 6.68% of analytes had a least one significant score (α=0.05) in 2014 compared to 10.29% in 2015. The number of tentatively identifiable, non-contaminant compounds in 2014 was 13 and 32 in 2015. A few tentatively identified compounds had ambiguous fragmentation, resulting in subsequent use of generic names (i.e. saturated hydrocarbon 1) in this study. Various hydrocarbon, ether, ester, aldehyde, ketone, halogenic, nitrogenous, and thioether compounds were tentatively identified (Table 4.1; Table 4.2). Furfural, 1-octen-3-ol, dimethyl disulfide, 3-carene, 2-isopropyl-3-methoxypyrazine, 2-phenylacetaldehyde, and α-copaene spectra were confirmed by standard retention time matching. A large number of tentatively identified aldehydes supports other assays that identified a large proportion of potato volatiles as aldehydes. Sensory analysis is necessary to verify tentatively identified or confirmed compounds that play a role in potato flavor.

A multitude of reasons may have contributed to a larger number of identifiable, variable compounds in 2015 versus 2014. Samples were stored for a longer period in 2014 prior to analysis, which may have caused sample degradation. Differences may also be attributable to systematic error within the instrument or cooking methods across years. User error is another potential factor, since technique and consistency tends to improve over time. Despite consistent production management, minor environmental differences in terms of water, light, nutrients, or other biotic factors may have also influenced compound presence and variation.

4.3.2. Spearman's Rank Correlations Between Potential Flavor Compounds

Cluster 1 is the most prominent, largest cluster ($0.67 \leq r_s \leq 0.95$) in the Spearman's rank correlation HCA heatmap, which includes (E)-2-heptenal (AD-14-1), 3,5-octadien-2-one (KE-14-2), o-methylacetophenone (KE-14-1), (E,E)-2,4-nonadiene (HC-14-1), 2-n-butyl furan (FU-14-1), 2,4-nonadienal (AD-14-3), (E)-2-octenal (AD-14-4), hexanal (AD-14-2), and 3,5-dimethylcyclopentene (HC-14-2) (Figure 4.1). The second largest cluster ($0.45 \leq r_s \leq 0.91$), cluster 5, contains dimethyl disulfide (SU-15), (E)-2-heptenal (AD-15-2), 1-nonanal (AD-15-3), benzoate-3-methyl-2-buten-1-ol (PA-15-1), benzaldehyde (AD-15-1), (Z)-2-methyl-2-penten-1-

ol (AC-15-3), and pentanal (AD-15-4). Two medium clusters contain 3-carene (TE-15-1), 2-isopropyl-3-methoxypyrazine (PZ-15), an unidentifiable phenol (AC-15-4), α-copaene (TE-15-2), and an unidentifiable phenyl acetate (PA-15-2) in cluster 6 ($0.43 \leq r_s \leq 0.98$) and unsaturated hydrocarbon 2 (HC-15-7), saturated hydrocarbon 2 (HC-15-4), saturated hydrocarbon 1 (HC-15-3), saturated hydrocarbon 3 (HC-15-5), and 2,2,3,4-tetramethyl-pentane (HC-15-2) in cluster 9 ($0.89 \leq r_s \leq 0.97$). Two weaker medium clusters are clusters 2 and 4, which include isomenthone (TE-15-3), 3-methyl-pentane (HC-15-1), unsaturated hydrocarbon 1 (HC-15-6), 1-octen-3-ol (AC-15-1), and p-methylacetophenone (KE-15-2) ($0.07 \leq r_s \leq 0.54$) and α-copaene (TE-14), methional (SU-14), 2-phenylacetaldehyde (AD-14-5), and furfural (FU-15-1) ($0.06 \leq r_s \leq 0.64$), respectively. Small clusters are as follows: 3,4,5-trimethyl-2-cyclopenten-1-one (KE-15-3), 1-pentanol (AC-15-2), and 2-ethylfuran (FU-15-1) ($0.58 \leq r_s \leq 0.69$) in cluster 3; 2-pentyl-furan (FU-15-2), 1-chloro-2-methyl-butane (HA-15), and 5-methyl-2-hexanone (KE-15-1) ($0.43 \leq r_s \leq 0.53$) in cluster 7; 3,4-dimethyl-styrene (HC-15-9) with azulene (HC-15-8) (r_s=0.66) in cluster 8.

Although clusters are evident in HCA of Spearman's rank correlation coefficients by analyte-to-analyte, the basis of the clustering is unclear. In general, compounds separate by sample years. Aldehydes occur in the two larger clusters, clusters 1 and 5, rather than separate across clusters as compounds with other functional groups (i.e. ketones, alcohols, hydrocarbons, etc.), perhaps due to a larger proportion of aldehydes relative to the other functional groups. Cluster 6 contains an alcohol, ester, pyrzaine, and two terpenes, which all have large atomic masses relative to the other compounds, though not all large mass compounds are within the cluster. Cluster 9 includes five hydrocarbons with retention times within a three-minute time window. While this study did not include characterizations of metabolite or product reactions for tentative or confirmed compounds, clustering between compounds could represent similar metabolite reactants or reactions.

4.3.3. ANOVA: Potential Flavor Compound Differences by Clone and Cooking Method

Univariate analysis revealed analyte variation and mean differences by clone, cooking method, and their interaction. Within 2014 data by clone, Russet Nugget and Yukon Gold have consistently larger mean peak areas than other clones, which is not evident for 2015 (Tables 4.3 & 4.4; Figures 4.2 & 4.3), indicating systematic or random errors as formerly mentioned. White-fleshed clone means were typically greater than purple- or red-fleshed clone means from 2014, while yellow-fleshed clone means were less distinct. Clone means in 2015 did not follow a similar pattern. For both years, analyte means are greater in baked versus boiled potatoes (Table 4.5; Figure 4.4). Clone by cooking method interactions are significant for several compounds across nine clones (Table 4.6).

Analyte mean differences by cooking method are more prominent in samples from 2015 than 2014, though all means for baked potatoes are greater than boiled. Intuitively, it makes sense that means are greater for baked potatoes since aluminum foil confined volatiles that may have been released during boiling. Analyte mean discrepancies between cooking methods are limited to nine of the fifteen clones. All white-fleshed clones have mean differences by cooking method for at least one analyte, while only some yellow-fleshed clones and no purple- or red-fleshed clones have differences. The most common compounds with mean differences by clone between cooking methods are aldehydes (including furans), suggesting formation dependence on cooking, most likely through Maillard or Strecker reactions.

4.3.4. HCA: Similarities Between Potential Flavor Compounds and Clones

For both years, HCA elucidates trends between analytes and clones (Figures 4.5 & 4.6). Two primary analyte clusters emerge in 2014 HCA, where cluster 2 contains four of five aldehydes (not including furans) and cluster 1 contains a mixture of compounds by functional group. The 2015 clone-to-analyte HCA also splits analytes into two clusters. The clusters differ by a few compounds by functional group, where cluster 1 uniquely contains both terpenes and nitrogenous compounds and cluster 2 contains most of the non-terpene hydrocarbons and both furans. Both clusters contain alcohols, aldehydes, and ketones. Except for the clustering of aldehydes in the 2014 HCA, analyte clustering in HCA of both years does not coincide with clustering from HCA of Spearman's rank correlation coefficients. Differential clustering of compounds across HCAs was expected due to the use of analyte means by clone rather than bivariate correlations between analytes

In 2014 HCA, white-fleshed clones cluster together in cluster B with the exception of Russet Nugget, which clusters with yellow-fleshed Yukon Gold in cluster A. Advanced selection CO04056-3P/PW and Purple Majesty, both purple-fleshed, correspond in cluster C. Cluster D is a mixture of yellow- and red-fleshed clones. Clustering by clone in 2015 HCA is more mixed than the 2014 HCA, though both purple-fleshed clones cluster together in cluster A similarly to the 2014 HCA. Contrasting clone clustering between years was anticipated because of differences in tentative or confirmed analyte identification. Clustering of clones in the 2014 analyte-to-clone HCA supports ANOVA differentiation of white- from colored-flesh clones, with the exception of Russet Nugget.

4.3.5. PCA: Potential Flavor Compounds in Relation to Clones

A PCA of 2014 data has four components that contribute to 92.2% of sample variation, whereas PCA of 2015 data has five components representing 71.2% of variation. Combined PCA for both years has six components expressing 82.4% of variation. Scores do not have well-defined separation by clone, cooking method, replication, year, or flesh color, however, some clustering occurs between compound loadings. Except where noted, compound clustering does not reflect characteristics including retention time, atomic mass, density, boiling point, vapor pressure, or carbon content.

In the 2014 model, there is an obvious small and large cluster of loadings, though the two clusters do not correlate well (Figure 4.7). Cluster 1 includes 2-phenylacetaldehyde (FU-14), furfural (AD-14-5), and methional (SU-14) and cluster 2 includes (E,E)-2,4-nonadiene (HC-14-1), 3,5-dimethylcyclopentene (HC-14-2), (E)-2-heptenal (AD-14-1), hexanal (AD-14-2), 2,4-nonadienal (AD-14-3), (E)-2-octenal (AD-14-4), o-methylacetophenone (KE-14-1), 3,5-octadien-2-one (KE-14-2), and 2-n-butyl furan (FU-14-1). Scores for Russet Nugget and Yukon Gold correlate positively to cluster 1, whereas purple- or red-fleshed scores appear to have a weak negative correlation to the same cluster.

For 2015, cluster 1 contains α-copaene (TE-15-2), an unidentifiable phenol (AC-15-4), and an unidentifiable phenyl acetate (PA-15-2) and correlates to cluster 2, which contains 2-isopropyl-3-methoxypyrazine (PZ-15), 3-carene (TE-15-1), p-methylacetophenone (KE-15-2), azulene (HC-15-8), and dimethyl disulfide (SU-15). Clusters 1 and 2 also have a weak correlation to hydrocarbon cluster 3 of 2,2,3,4-tetramethyl-pentane (HC-15-2), saturated hydrocarbons 1 to 3 (HC-15-3, HC-15-4, HC-15-5), and unsaturated hydrocarbon 2 (HC-15-7) (Figure 4.8). The hydrocarbons weakly correlate to cluster 4, which contains most of the aldehydes and ketones, including 1-pentanol (AC-15-2), 1-nonanal (AD-15-3), (E)-2-heptenal (AD-15-2), pentanal (AD-15-4), 5-methyl-2-hexanone (KE-15-1), 3,4,5-trimethyl-2-cyclopenten-1-one (KE-15-3), 2-ethylfuran (FU-15-1), 2-pentyl-furan (FU-15-2), and 1-chloro-2-methyl-butane (HA-15). Cluster 4 is also related to baked Russet Nugget scores. The former cluster also appears to be somewhat positively related to other white-fleshed clone scores and negatively to red-fleshed clone scores. In both PCAs by year, white- and purple- or red-fleshed clone means appear to be in opposition.

4.3.6. Post-Hoc ANOVA: Potential Flavor Compound Differences by Flesh Color

Although 2015 HCA of clones and compounds does not have clear trends, post-hoc ANOVA of compounds by flesh color suggests otherwise. For 2014 samples, compounds with significant variation across flesh color generally have a greater mean for white-fleshed clones, followed by yellow- and purple- or red-fleshed clones (Table 4.7; Figure 4.9). Compound means are partially greater for white- and purple- or red-fleshed clones in 2015 samples (Table 4.8; Figure 4.10), though differences between all three flesh color groups are less distinct as in 2014. Compounds with different means across flesh color include diverse primary functional groups. Hydrocarbon content is greater for white- versus purple- or red-fleshed potatoes in 2014 samples, while the opposite is true for 2015 samples. Yellow-fleshed potato hydrocarbon means in either year are similar to both purple- or red- and white-fleshed clone means. Aldehyde means across both years tend to be greatest in white- followed by yellow-, and purple- or red- potatoes. Ketone means in both years are greater for white- relative to purple- or red-fleshed clones.

4.3.7. Application of Results

Sample runtimes for HS-SPME GC-MS were relatively long for practical use in high-throughput phenotyping, but methods could be streamlined to improve feasibility for application in selection-based breeding programs. Univariate mean differences of potential flavor compounds between clones suggest diverse cooked potato flavor profiles with a possible genetic component. Multivariate flavor profiles by clone from HCA and PCA are less diverse and more inconsistent compared to univariate flavor profiles by clone, though some trends occur by flesh color. Flesh color is somewhat related to genetic background, particularly for white- versus colored-fleshed potato clones, so a genetic component for flavor profiles is still plausible. Flavor phenotyping and selection may be possible by selecting a few, key flavor compounds as biomarkers. Nonetheless, the role of compounds in flavor should be confirmed with sensory analysis prior to breeding or other genetic explorations. Cooking methods result in differences in relative volatile amounts, which may influence potato flavor.

4.4. Summary

Cooked fresh market potato volatile and semi-volatile diversity was examined. Volatiles are aroma compounds that influence flavor to a greater extent than non-volatile, taste compounds. To evaluate potential key aroma compounds for potato flavor, headspace profiles of 15 clones with a range of skin and flesh colors were compared. Across 2014 and 2015, 33 compounds with differences by clone, cooking method, or interactions (ANOVA F-test and Tukey's HSD t-tests, $\alpha=0.05$, n=5 technical replicates x clone x cooking method) were tentatively identified using mass spectral matching. Identification of 8 compounds was confirmed by retention time matching in gas chromatography, including 3-carene, α-copaene, dimethyl disulfide, furfural, methional, 1-octen-3-ol, 2-phenacetaldehyde, and 2-isopropyl-3-methoxypyrazine. Clone differences revealed some compound trends by flesh color, particularly for white versus other flesh colors. Compound peak area means were greater for baked versus boiled potatoes, suggesting aroma flavor differences between cooking methods.

Table 4.1. Characteristics of analytes detected by HS-SPME GC-MS with spectral matching identification[a]

Year	Primary Functional Group[b]	Configuration	Compound	Organoleptics[c]	Significant Variable[d]	Compound Code[e]
2014	**Hydrocarbon**	**Alkene**	(E,E)-2,4-nonadiene		cL	HC-14-1
		Cycloalkene	3,5-dimethylcyclopentene		cL, cL:cK	HC-14-2
		Terpene	α-copaene*	woody, spicy, honey	cL	TE-14
	Aldehyde	**Aliphatic**	(E)-2-heptenal	green, fatty, fruity	cL, cL:cK	AD-14-1
			hexanal	green, woody, fatty	cL, cL:cK	AD-14-2
			2,4-nonadienal	fatty, nutty, citrus	cL, cL:cK	AD-14-3
			(E)-2-octenal	green, herbal, fatty	cL, cL:cK	AD-14-4
			2-phenylacetaldehyde*	honey, floral, cocoa	cL, cK	AD-14-5
	Ketone	**Aliphatic**	o-methylacetophenone	floral, burnt, nutty	cL	KE-14-1
			3,5-octadien-2-one	fruity, green	cL	KE-14-2
	Ether	**Furan**	2-n-butyl furan	fruity, wine, spicy	cL, cL:cK	FU-14-1
			furfural*	caramel, woody, nutty	cK	FU-14-2
	Thioether	**Aliphatic with Aldehyde**	methional*	potato, musty, fatty	cK	SU-14
2015	**Hydrocarbon**	**Alkane**	3-methyl-pentane		cL, cL:cK, F	HC-15-1
			2,2,3,4-tetramethyl-pentane		cL, cK, cL:cK	HC-15-2
			saturated hydrocarbon 1		cL, cK	HC-15-3
			saturated hydrocarbon 2		cL, cK, cL:cK	HC-15-4
			saturated hydrocarbon 3		cL, cK	HC-15-5
		Alkene	unsaturated hydrocarbon 1		cL, cL:cK, F	HC-15-6
			unsaturated hydrocarbon 2		cL, cK, F	HC-15-7
		Cycloalkene	azulene		cK	HC-15-8
		Styrene	3,4-dimethyl-styrene		cK	HC-15-9
		Terpene	3-carene*	citrus, pine	cL, cL:cK	TE-15-1
			α-copaene*	woody, spicy, honey	cL, cL:cK	TE-15-2
			isomenthone	minty	cK	TE-15-3
	Alcohol	**Aliphatic**	1-octen-3-ol*	earthy, green, fatty	cL, cK, cL:cK	AC-15-1
			1-pentanol	fermented, yeasty, wine	cL, cL:cK, F	AC-15-2
			(Z)-2-methyl-2-penten-1-ol		cL, cK, F	AC-15-3
		Phenol	unidentifiable phenol		cL:cK	AC-15-4
	Aldehyde	**Aliphatic**	benzaldehyde	nutty, fruity, fatty	cL, cL:cK, F	AD-15-1
			(E)-2-heptenal	green, fatty, fruity	cL, cK, cL:cK, F	AD-15-2
			1-nonanal	citrus, green, potato	cL, cL:cK	AD-15-3
			pentanal	fermented, yeasty, wine	cL, cL:cK, F	AD-15-4
	Ketone	**Aliphatic**	5-methyl-2-hexanone		cL, cK, cL:cK, F	KE-15-1
		Acetophenone	p-methylacetophenone	creamy, fruity, vanilla	cL, cK, cL:cK	KE-15-2
		Cyclic	3,4,5-trimethyl-2-cyclopenten-1-one		cL, cL:cK, F	KE-15-3
	Ether	**Furan**	2-ethylfuran	musty, earthy, yeasty	cL, cK, cL:cK, F	FU-15-1
			2-pentyl-furan	fruity, green, earthy	cL, cK, cL:cK, F	FU-15-2
			furfural*	caramel, woody, nutty	cK, cL:cK	FU-15-3
	Ester	**Phenyl Acetate**	benzoate-3-methyl-2-buten-1-ol	woody, fruity, chocolate	cL	PA-15-1
		Phenyl Acetate	unidentifiable phenyl acetate		cL	PA-15-2
	Halogenic	**Aliphatic**	1-chloro-2-methyl-butane		cK, cL:cK	HA-15
	Nitrogenous	**Pyrazine**	2-isopropyl-3-methoxypyrazine*	earthy, chocolate, nutty	cL, cL:cK, F	PZ-15
		Pyrrole	2-formyl-1-methylpyrrole		cL, cL:cK, F	PR-15
	Thioether	**Aliphatic**	dimethyl disulfide*	sulfur, cabbage, caramel	cL:cK	SU-15

[a]Spectral matching with National Institute of Standards and Technology (NIST, Gaithersburg, MD) 11 database and NIST MS Search Software version 2.0f, minimum of 450 matching intensity for positive identification. *An asterisks indicates that analyte identification was additionally confirmed using retention time matching of a standard.
[b]Primary functional groups based on flavor metabolites.
[c]Top 3 orrganoleptic characteristics retrieved from the Good Scents Company online database (Oak Creek, WI). Nutty includes almond, green includes leafy, vegetative, and grassy, fatty includes buttery, chicken fat, oily, beef, and egg.
[d]Analytes have significant variation according to ANOVA F-tests (α=0.05) for cL=clone, cK=cooking method, cL:cK=clone by cooking method, or F=flesh color as notated.
[e]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.

Table 4.2. Top 5 mass peaks used in spectral matching[a] and retention times of cooked potato analytes

Compound	Top 5 Mass Peaks	RT
unsaturated hydrocarbon 1	57, 56, 55, 53, 61	144
1-chloro-2-methyl-butane	57, 41, 55, 39, 56	158
2-ethylfuran	81, 53, 96, 65, 82	171
pentanal	44, 58, 43, 41, 57	172
3-methyl-pentane	57, 56, 53, 50, 51	226
1-pentanol	41, 55, 42, 71, 70	276
(Z)-2-methyl-2-penten-1-ol	72, 71, 67, 43 57	332
hexanal	41, 44, 56, 43, 57	348
furfural* (2015)	95, 97, 39, 69, 68	395
furfural* (2014)	96, 95, 39, 38, 37	418
3,4,5-trimethyl-2-cyclopenten-1-one	109, 81, 79, 124, 53	428
5-methyl-2-hexanone	43, 58, 81, 71, 82	503
2-formyl-1-methylpyrrole	108, 109, 42, 40, 95	546
methional	48, 49, 30, 50, 35	559
(E,E)-2,4-nonadiene	79, 81, 53, 95, 68	624
(E)-2-heptenal (2015)	41, 55, 57, 39, 69	632
benzaldehyde	77, 106, 51, 50, 52	642
(E)-2-heptenal (2014)	41, 83, 55, 39, 69	649
1-octen-3-ol*	57, 69, 55, 67, 81	674
2-pentyl-furan	82, 81, 53, 138, 39	686
benzoate-3-methyl-2-buten-1-ol	105, 41, 43, 55, 69	716
dimethyl disulfide*	93, 91, 79, 136, 81	716
3-carene*	93, 67, 121, 81, 53	732
3,5-dimethylcyclopentene	81, 79, 41, 53, 39	749
saturated hydrocarbon 1	57, 41, 43, 71, 70	758
saturated hydrocarbon 2	57, 41, 56, 71, 70	795
2-phenylacetaldehyde	91, 92, 65, 93, 63	810
2,2,3,4-tetramethyl-pentane	57, 41, 43, 56, 71	818
(E)-2-octenal	41, 55, 83, 70, 39	839
unsaturated hydrocarbon 2	57, 41, 56, 55, 69	845
2-isopropyl-3-methoxypyrazine*	109, 152, 137, 124, 105	883
1-nonanal	41, 57, 43, 55, 56	912
3,5-octadien-2-one	95, 79, 81, 71, 59	914
saturated hydrocarbon 3	57, 41, 56, 71, 68	934
p-methylacetophenone	91, 119, 92, 134, 65	936
3,4-dimethyl-styrene	117, 119, 115, 68, 132	1002
isomenthone	55, 69, 41, 112, 70	1023
o-methylacetophenone	91, 79, 105, 134, 119	1063
azulene	128, 129, 102, 127, 126	1084
2,4-nonadienal	81, 41, 67, 39, 65	1172
2-n-butyl furan	81, 67, 95, 70, 82	1363
phenyl with ester branch	105, 91, 79, 93, 106	1435
α-copaene* (2015)	161, 119, 149, 204, 145	1437
unidentifiable phenol	148, 133, 147, 103, 129	1439
α-copaene* (2014)	105, 119, 91, 161, 93	1452

[a]Spectral matching with National Institute of Standards and Technology (NIST, Gaithersburg, MD) 11 database and NIST MS Search Software version 2.0f, minimum of 450 matching intensity for positive identification.

*An asterisks indicates that analyte identification was additionally confirmed using retention time matching of a standard.

Table 4.3. Log-transformed peak area clone means and comparisons[a] and 95% confidence intervals of analytes detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014[b]

Flesh Color	Clone	(E,E)-2,4-nonadiene	3,5-dimethylcyclo-pentene	α-copaene	(E)-2-heptenal	hexanal	2,4-nonadienal	(E)-2-octenal	2-phenyl-acetaldehyde
White	**AC00395-2RU**	1.93 [1.71, 2.14] AC	2.34 [2.07, 2.60] AC	1.79 [1.68, 1.90] AB	2.50 [2.31, 2.69] AC	3.17 [2.89, 3.45] AD	2.21 [2.05, 2.36] AB	2.24 [2.06, 2.43] AD	2.48 [2.30, 2.67] AB
	Fortress Russet	1.73 [1.57, 1.89] AB	2.16 [1.88, 2.43] AB	2.16 [1.98, 2.34] BC	2.40 [2.19, 2.61] AC	2.90 [2.61, 3.19] ABC	2.08 [1.94, 2.21] A	2.09 [1.91, 2.28] ABC	2.34 [2.18, 2.50] AB
	Russet Burbank	1.84 [1.69, 1.98] AC	2.30 [2.04, 2.56] AC	2.16 [2.04, 2.27] BC	2.49 [2.30, 2.69] AC	3.30 [2.90, 3.69] BD	2.27 [2.04, 2.49] AB	2.27 [2.04, 2.49] BD	2.67 [2.50, 2.85] B
	Russet Norkotah	1.66 [1.56, 1.77] AB	2.15 [1.98, 2.33] AB	1.74 [1.50, 1.98] A	2.42 [2.29, 2.55] AC	2.94 [2.74, 3.13] ABC	2.13 [1.97, 2.29] A	2.11 [1.98, 2.24] AD	2.47 [2.33, 2.61] AB
	Russet Nugget	2.24 [1.97, 2.51] C	2.79 [2.48, 3.10] C	1.84 [1.73, 1.95] AB	2.75 [2.58, 2.92] C	3.72 [3.37, 4.07] D	2.58 [2.37, 2.78] B	2.51 [2.32, 2.71] D	2.60 [2.44, 2.76] AB
Yellow	**CO04067-8R/Y**	1.68 [1.55, 1.81] AB	1.97 [1.77, 2.16] A	1.88 [1.72, 2.03] AC	2.09 [1.92, 2.26] A	2.66 [2.42, 2.90] AB	1.97 [1.84, 2.10] A	1.91 [1.76, 2.05] AB	2.62 [2.35, 2.88] AB
	CO04099-3W/Y	1.69 [1.53, 1.86] AB	2.01 [1.83, 2.19] A	1.94 [1.72, 2.16] AC	2.21 [2.10, 2.32] AB	2.65 [2.45, 2.84] AB	1.96 [1.83, 2.10] A	1.95 [1.79, 2.11] AB	2.55 [2.38, 2.73] AB
	Harvest Moon	1.66 [1.49, 1.82] AB	1.90 [1.74, 2.06] A	1.72 [1.60, 1.84] A	2.13 [2.03, 2.23] A	2.56 [2.33, 2.80] AB	1.96 [1.86, 2.07] A	1.90 [1.74, 2.07] AB	2.70 [2.55, 2.85] B
	Masquerade	1.70 [1.50, 1.89] AB	2.03 [1.79, 2.28] A	1.94 [1.76, 2.12] AC	2.30 [2.12, 2.47] AB	2.76 [2.39, 3.13] AB	2.05 [1.87, 2.24] A	2.00 [1.82, 2.18] ABC	2.58 [2.45, 2.72] AB
	Red Luna	1.74 [1.57, 1.91] AB	2.23 [1.90, 2.55] AB	1.80 [1.70, 1.90] AB	2.38 [2.19, 2.57] AC	2.99 [2.57, 3.41] ABC	2.13 [1.93, 2.32] A	2.08 [1.87, 2.29] ABC	2.66 [2.52, 2.81] B
	Yukon Gold	2.04 [1.80, 2.29] BC	2.61 [2.28, 2.94] BC	1.91 [1.77, 2.06] AC	2.58 [2.34, 2.82] BC	3.56 [3.15, 3.97] CD	2.53 [2.31, 2.75] B	2.40 [2.16, 2.65] CD	2.63 [2.45, 2.80] AB
Purple	**CO04056-3P/PW**	1.68 [1.59, 1.78] AB	1.87 [1.77, 1.98] A	2.25 [2.06, 2.43] C	2.09 [1.94, 2.24] A	2.58 [2.39, 2.78] AB	1.90 [1.81, 2.00] A	1.89 [1.78, 1.99] AB	2.31 [2.17, 2.46] AB
	Purple Majesty	1.60 [1.50, 1.70] A	1.83 [1.73, 1.93] A	2.14 [1.99, 2.30] BC	2.17 [2.00, 2.33] AB	2.51 [2.38, 2.64] A	1.91 [1.82, 1.99] A	1.83 [1.72, 1.94] A	2.24 [2.10, 2.39] A
Red	**CO04063-4R/R**	1.63 [1.52, 1.74] AB	1.87 [1.74, 2.01] A	1.73 [1.61, 1.85] A	2.06 [1.93, 2.20] A	2.56 [2.35, 2.78] AB	1.92 [1.80, 2.04] A	1.85 [1.72, 1.97] AB	2.45 [2.28, 2.62] AB
	Crimson King	1.63 [1.54, 1.72] A	1.90 [1.79, 2.02] A	1.66 [1.54, 1.78] A	2.13 [2.00, 2.26] A	2.74 [2.51, 2.98] AB	1.95 [1.85, 2.05] A	1.93 [1.80, 2.06] AB	2.36 [2.17, 2.55] AB
	Compound Code	*HC-14-1*	*HC-14-2*	*TE-14*	*AD-14-1*	*AD-14-2*	*AD-14-3*	*AD-14-4*	*AD-14-5*
	Configuration	*Alkene*	*Cycoalkene*	*Terpene*			*Aliphatic*		
	Functional Group		*Hydrocarbon*				*Aldehyde*		

Flesh Color	Clone	o-methylaceto-phenone	3,5-octadien-2-one	2-n-butyl furan
White	**AC00395-2RU**	1.96 [1.80, 2.12] AC	1.91 [1.69, 2.13] AC	1.90 [1.74, 2.05] AC
	Fortress Russet	1.88 [1.72, 2.04] AB	1.70 [1.54, 1.86] AB	1.85 [1.67, 2.03] AC
	Russet Burbank	1.98 [1.83, 2.12] AC	1.87 [1.73, 2.02] AC	1.94 [1.72, 2.16] AC
	Russet Norkotah	1.77 [1.67, 1.88] A	1.72 [1.60, 1.83] AB	1.78 [1.63, 1.93] AB
	Russet Nugget	2.34 [2.09, 2.59] C	2.26 [2.03, 2.49] C	2.21 [2.00, 2.43] C
Yellow	**CO04067-8R/Y**	1.84 [1.67, 2.01] AB	1.72 [1.59, 1.86] AB	1.67 [1.48, 1.85] A
	CO04099-3W/Y	1.80 [1.63, 1.97] AB	1.72 [1.53, 1.90] AB	1.69 [1.54, 1.84] A
	Harvest Moon	1.84 [1.69, 1.99] AB	1.70 [1.56, 1.84] AB	1.63 [1.48, 1.79] A
	Masquerade	1.80 [1.61, 1.99] AB	1.70 [1.52, 1.87] AB	1.70 [1.53, 1.87] A
	Red Luna	1.89 [1.74, 2.05] AB	1.82 [1.64, 2.00] AB	1.76 [1.58, 1.95] AB
	Yukon Gold	2.18 [1.94, 2.43] BC	2.08 [1.81, 2.35] BC	2.15 [1.95, 2.36] BC
Purple	**CO04056-3P/PW**	1.80 [1.70, 1.90] AB	1.66 [1.55, 1.77] A	1.60 [1.51, 1.69] A
	Purple Majesty	1.76 [1.66, 1.87] A	1.66 [1.55, 1.76] A	1.59 [1.49, 1.70] A
Red	**CO04063-4R/R**	1.81 [1.67, 1.94] AB	1.70 [1.56, 1.84] AB	1.65 [1.51, 1.78] A
	Crimson King	1.81 [1.66, 1.96] AB	1.71 [1.61, 1.80] AB	1.68 [1.57, 1.79] A
	Compound Code	*KE-14-1*	*KE-14-2*	*FU-14-2*
	Configuration	*Aliphatic*		*Furan*
	Functional Group	*Ketone*		*Ether*

[a]Clone means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05) (n=technical replicates x 15 clones).

[b]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.

Table 4.4. Log-transformed peak area clone means and comparisons[a] with 95% confidence intervals of analytes detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2015[b]

Flesh	Clone	3-methyl-pentane	2,2,3,4-tetramethyl-pentane	saturated hydrocarbon 1	saturated hydrocarbon 2	saturated hydrocarbon 3	unsaturated hydrocarbon 1	unsaturated hydrocarbon 2	3-carene	α-copaene
White	AC00395-2RU	3.33 [3.20, 3.46] AC	3.53 [3.47, 3.60] CD	3.04 [3.00, 3.09] C	3.47 [3.39, 3.54] BD	3.54 [3.46, 3.62] BC	3.70 [3.54, 3.86] AB	3.30 [3.25, 3.35] BC	1.98 [1.91, 2.04] AB	2.58 [2.42, 2.74] AC
	Fortress Russet	3.40 [3.33, 3.48] CEF	3.15 [3.04, 3.26] AD	2.80 [2.69, 2.92] AC	3.26 [3.19, 3.34] AD	3.11 [2.99, 3.22] AB	3.61 [3.47, 3.76] BC	3.10 [3.02, 3.18] AC	2.20 [2.14, 2.26] F	3.16 [2.98, 3.33] EF
	Russet Burbank	3.90 [3.78, 4.02] CEF	3.32 [3.23, 3.40] ABC	2.92 [2.83, 3.01] AB	3.34 [3.29, 3.39] ABC	3.28 [3.18, 3.37] AC	3.96 [3.83, 4.09] AB	3.16 [3.10, 3.23] A	2.38 [2.33, 2.42] A	3.05 [2.94, 3.17] CE
	Russet Norkotah	3.86 [3.78, 3.93] A	3.21 [3.10, 3.32] ABC	2.88 [2.78, 2.98] A	3.21 [3.14, 3.27] ABC	3.12 [3.00, 3.23] A	3.97 [3.77, 4.18] AB	3.07 [2.99, 3.14] AB	2.00 [1.95, 2.05] AC	2.29 [2.22, 2.37] CF
	Russet Nugget	3.25 [3.13, 3.38] DEG	3.41 [3.35, 3.47] ABC	2.94 [2.88, 3.00] AB	3.38 [3.31, 3.45] ABC	3.38 [3.29, 3.46] AC	3.83 [3.71, 3.96] BC	3.18 [3.11, 3.24] AC	2.16 [2.03, 2.29] A	2.79 [2.59, 2.99] A
Yellow	CO04067-8R/Y	3.60 [3.48, 3.72] FG	3.50 [3.37, 3.63] ABC	3.04 [2.94, 3.14] AC	3.48 [3.38, 3.58] ABC	3.50 [3.36, 3.64] C	3.89 [3.74, 4.04] AC	3.26 [3.17, 3.35] AC	2.23 [2.18, 2.28] EF	3.06 [2.95, 3.18] DEF
	CO04099-3W/Y	3.84 [3.69, 3.99] BC	3.27 [3.10, 3.44] A	2.93 [2.80, 3.07] A	3.24 [3.14, 3.34] A	3.21 [3.03, 3.38] A	3.72 [3.51, 3.93] A	3.14 [3.02, 3.25] AB	2.49 [2.41, 2.57] CE	2.96 [2.77, 3.16] F
	Harvest Moon	3.56 [3.44, 3.68] EG	3.23 [3.11, 3.36] ABC	2.85 [2.73, 2.97] AB	3.26 [3.18, 3.34] ABC	3.16 [3.02, 3.30] AB	3.93 [3.85, 4.02] AB	3.10 [3.01, 3.20] AC	1.97 [1.88, 2.06] F	2.20 [2.02, 2.38] CF
	Masquerade	3.05 [2.91, 3.18] CEF	3.25 [3.10, 3.40] BD	2.83 [2.70, 2.95] BC	3.25 [3.11, 3.39] CD	3.17 [3.01, 3.34] A	3.81 [3.64, 3.99] AB	3.09 [2.97, 3.21] AC	2.00 [1.85, 2.14] DE	2.89 [2.68, 3.10] DEF
	Red Luna	3.52 [3.43, 3.61] BCD	3.29 [3.17, 3.40] ABC	2.94 [2.85, 3.03] AB	3.29 [3.22, 3.36] ABC	3.22 [3.10, 3.35] AC	3.93 [3.77, 4.08] AC	3.10 [3.01, 3.18] AB	1.99 [1.92, 2.06] AB	2.66 [2.51, 2.80] BCD
	Yukon Gold	3.64 [3.41, 3.86] G	3.28 [3.14, 3.42] D	2.89 [2.80, 2.99] C	3.24 [3.14, 3.34] D	3.22 [3.08, 3.36] A	3.88 [3.76, 4.01] C	3.07 [2.97, 3.18] C	1.95 [1.86, 2.04] ACD	2.73 [2.58, 2.88] CF
Purple	CO04056-3P/PW	3.59 [3.50, 3.67] EG	3.34 [3.24, 3.44] A	2.97 [2.89, 3.05] A	3.31 [3.23, 3.38] AB	3.30 [3.19, 3.40] AC	3.98 [3.93, 4.04] BC	3.17 [3.09, 3.25] A	2.46 [2.41, 2.52] AC	3.07 [2.90, 3.24] AB
	Purple Majesty	3.97 [3.88, 4.06] BCE	3.60 [3.51, 3.69] AB	3.11 [3.06, 3.16] A	3.49 [3.40, 3.58] AB	3.57 [3.47, 3.68] C	4.27 [4.20, 4.34] AC	3.34 [3.27, 3.40] AB	2.11 [2.01, 2.21] AB	2.89 [2.74, 3.03] A
Red	Crimson King	3.81 [3.66, 3.95] AB	3.29 [3.20, 3.38] AD	2.93 [2.85, 3.01] AC	3.27 [3.20, 3.34] AD	3.23 [3.12, 3.34] AB	4.00 [3.87, 4.14] AB	3.14 [3.09, 3.20] AC	1.95 [1.90, 1.99] BCD	2.24 [2.01, 2.47] CF
	Compound Code	*HC-15-1*	*HC-15-2*	*HC-15-3*	*HC-15-4*	*HC-15-5*	*HC-15-6*	*HC-15-7*	*TE-15-1*	*TE-15-2*
	Configuration			*Alkane*			*Alkene*		*Terpene*	
	Functional Group					*Hydrocarbon*				

Flesh	Clone	1-octen-3-ol	1-pentanol	(Z)-2-methyl-2-penten-1-ol	benzaldehyde	(E)-2-heptenal	nonanal	pentanal	5-methyl-2-hexanone	p-methylaceto-phenone	3,4,5-trimethyl-2-cyclopenten-1-one
White	AC00395-2RU	3.25 [3.16, 3.35] CDE	3.50 [3.16, 3.85] C	3.36 [3.23, 3.50] AB	3.37 [3.23, 3.51] AB	2.53 [2.47, 2.59] AB	3.29 [3.11, 3.46] AC	3.65 [3.52, 3.77] ABC	3.08 [2.86, 3.31] BC	2.90 [2.84, 2.97] ABC	3.21 [2.99, 3.42] CDF
	Fortress Russet	3.14 [3.05, 3.23] BD	3.30 [3.09, 3.51] AB	3.14 [3.04, 3.24] AB	3.20 [3.04, 3.35] AC	2.43 [2.34, 2.53] A	3.31 [3.12, 3.50] AC	3.40 [3.18, 3.61] AB	2.95 [2.84, 3.06] AC	2.91 [2.86, 2.96] AB	3.36 [3.18, 3.53] A
	Russet Burbank	3.21 [3.18, 3.25] EF	3.08 [2.99, 3.16] AB	3.53 [3.46, 3.59] AB	3.55 [3.50, 3.60] CD	2.65 [2.58, 2.72] AB	3.57 [3.51, 3.63] AC	3.52 [3.31, 3.72] AB	2.85 [2.64, 3.06] AC	3.19 [3.10, 3.27] ABC	3.14 [2.99, 3.29] BCDF
	Russet Norkotah	3.03 [2.93, 3.14] AB	3.34 [3.07, 3.62] BC	3.40 [3.30, 3.50] AB	3.47 [3.39, 3.55] CD	2.60 [2.53, 2.67] BC	3.43 [3.25, 3.61] C	3.64 [3.53, 3.74] CD	2.85 [2.73, 2.97] AB	2.90 [2.86, 2.94] AB	3.10 [2.93, 3.27] EF
	Russet Nugget	3.04 [2.93, 3.15] ABC	3.23 [3.00, 3.46] AC	4.49 [4.20, 4.78] A	3.66 [3.61, 3.71] A	2.84 [2.66, 3.02] A	3.46 [3.42, 3.51] AB	4.09 [3.85, 4.33] A	3.10 [2.84, 3.35] AC	2.77 [2.67, 2.86] ABC	3.49 [3.15, 3.84] ADE
Yellow	CO04067-8R/Y	2.97 [2.93, 3.02] BED	2.96 [2.85, 3.06] AC	3.55 [3.24, 3.86] B	3.44 [3.25, 3.63] BCD	2.49 [2.37, 2.62] AC	3.39 [3.27, 3.50] BC	3.61 [3.40, 3.83] AB	2.80 [2.72, 2.88] AC	2.87 [2.82, 2.92] D	2.93 [2.82, 3.04] BCDF
	CO04099-3W/Y	3.58 [3.50, 3.66] ABC	3.51 [3.20, 3.81] AC	3.07 [2.87, 3.26] AB	3.24 [3.08, 3.40] A	2.58 [2.48, 2.67] A	3.17 [2.96, 3.38] AC	3.25 [3.01, 3.49] AB	3.13 [2.90, 3.37] AC	3.07 [2.99, 3.15] ABC	3.31 [3.14, 3.48] DF
	Harvest Moon	3.04 [2.97, 3.11] F	3.09 [3.00, 3.18] C	3.45 [3.34, 3.56] A	3.75 [3.66, 3.84] A	2.54 [2.46, 2.61] AC	3.61 [3.49, 3.73] A	3.71 [3.56, 3.87] A	2.74 [2.66, 2.81] C	2.86 [2.78, 2.94] CD	3.07 [2.88, 3.27] CDF
	Masquerade	3.04 [2.94, 3.13] A	3.35 [3.25, 3.45] AB	3.46 [3.27, 3.65] B	3.68 [3.62, 3.75] AC	2.79 [2.55, 3.03] A	3.60 [3.48, 3.71] AC	4.06 [3.81, 4.30] ABC	2.73 [2.62, 2.84] AC	2.84 [2.67, 3.01] ABC	3.44 [3.28, 3.60] AC
	Red Luna	3.24 [3.15, 3.33] BD	3.04 [2.91, 3.17] AB	3.32 [3.19, 3.45] AB	3.55 [3.46, 3.64] BCD	2.48 [2.37, 2.60] A	3.53 [3.44, 3.62] BC	3.73 [3.61, 3.85] BD	2.71 [2.65, 2.77] A	2.99 [2.90, 3.08] BD	2.97 [2.82, 3.12] AD
	Yukon Gold	3.45 [3.33, 3.58] DF	2.93 [2.81, 3.06] A	3.36 [3.21, 3.50] AB	3.68 [3.58, 3.77] A	2.57 [2.49, 2.64] A	3.46 [3.36, 3.56] AC	3.37 [3.16, 3.59] AB	2.81 [2.65, 2.96] AC	2.90 [2.82, 2.97] BD	3.15 [3.01, 3.29] AB
Purple	CO04056-3P/PW	3.24 [3.16, 3.32] AB	2.98 [2.90, 3.05] BC	3.18 [3.12, 3.24] AB	3.41 [3.27, 3.55] AD	2.42 [2.37, 2.48] AC	3.48 [3.34, 3.62] AC	3.41 [3.35, 3.48] ABC	2.91 [2.85, 2.97] AC	2.86 [2.77, 2.95] ABC	2.65 [2.59, 2.71] BCDF
	Purple Majesty	3.39 [3.34, 3.44] AB	2.84 [2.77, 2.92] AC	3.29 [3.12, 3.46] AB	3.24 [3.07, 3.41] D	2.47 [2.39, 2.54] AB	3.29 [3.14, 3.44] C	3.50 [3.43, 3.56] BD	2.81 [2.76, 2.86] AB	2.99 [2.92, 3.05] AB	2.77 [2.70, 2.85] ADF
Red	Crimson King	3.09 [3.03, 3.15] AB	3.13 [2.95, 3.31] AC	3.07 [2.96, 3.19] C	3.17 [3.02, 3.31] BCD	2.39 [2.34, 2.44] C	3.24 [3.12, 3.36] AC	3.24 [3.07, 3.42] D	2.89 [2.83, 2.95] BC	2.89 [2.80, 2.97] A	3.04 [2.91, 3.16] F
	Compound Code	*AC-15-1*	*AC-15-2*	*AC-15-3*	*AD-15-1*	*AD-15-2*	*AD-15-3*	*AD-15-4*	*KE-15-1*	*KE-15-2*	*KE-15-3*
	Configuration		*Aliphatic*				*Aliphatic*		*Aliphatic*	*Acetophenone*	*Cyclic*
	Functional Group		*Alcohol*				*Aldehyde*			*Ketone*	

Flesh	Clone	2-ethylfuran	2-pentyl-furan	benzoate-3-methyl-2-buten-1-ol	unidentifiable phenyl acetate	2-Isopropyl-3-methoxypyrazine	2-formyl-1-methylpyrrole
White	AC00395-2RU	3.53 [3.10, 3.95] BC	3.40 [3.20, 3.61] BC	3.17 [3.09, 3.25] AC	2.25 [1.72, 2.78] AD	2.82 [2.68, 2.96] A	2.63 [2.54, 2.72] AB
	Fortress Russet	3.34 [2.97, 3.71] A	3.16 [3.00, 3.32] A	3.08 [2.95, 3.21] AC	2.60 [2.16, 3.04] D	2.82 [2.65, 2.99] B	2.96 [2.74, 3.19] AB
	Russet Burbank	2.97 [2.80, 3.13] A	3.17 [3.06, 3.27] A	3.35 [3.30, 3.41] AC	2.47 [2.03, 2.90] AD	2.74 [2.61, 2.87] A	2.72 [2.65, 2.80] AB
	Russet Norkotah	3.25 [2.80, 3.70] AC	3.08 [2.89, 3.28] A	3.24 [3.12, 3.36] C	2.17 [1.79, 2.56] CD	2.71 [2.63, 2.80] A	2.74 [2.70, 2.77] AB
	Russet Nugget	3.61 [3.25, 3.97] A	3.42 [3.17, 3.67] A	3.33 [3.27, 3.39] A	2.41 [1.83, 2.99] AB	2.70 [2.58, 2.83] A	2.64 [2.52, 2.76] A
Yellow	CO04067-8R/Y	3.02 [2.84, 3.20] AB	3.05 [2.93, 3.17] AC	3.19 [3.08, 3.29] C	2.51 [2.03, 2.99] D	2.71 [2.60, 2.83] A	2.55 [2.44, 2.66] AC
	CO04099-3W/Y	3.07 [2.80, 3.34] AC	3.10 [2.90, 3.31] AC	3.09 [2.93, 3.24] AB	2.47 [1.86, 3.08] D	2.78 [2.65, 2.92] A	2.60 [2.45, 2.76] C
	Harvest Moon	2.95 [2.81, 3.10] AC	3.08 [2.98, 3.17] AC	3.37 [3.28, 3.47] AB	2.12 [1.68, 2.57] D	2.77 [2.67, 2.87] A	2.83 [2.75, 2.91] AB
	Masquerade	3.04 [2.90, 3.18] AC	2.97 [2.89, 3.04] AB	3.38 [3.22, 3.55] AC	2.49 [1.98, 3.01] D	2.70 [2.58, 2.82] A	2.58 [2.49, 2.68] AB
	Red Luna	3.00 [2.88, 3.13] AC	2.88 [2.76, 2.99] A	3.28 [3.19, 3.37] AC	2.32 [1.90, 2.75] AD	2.67 [2.59, 2.75] A	2.62 [2.55, 2.70] AB
	Yukon Gold	2.84 [2.69, 2.98] A	2.86 [2.79, 2.92] AC	3.26 [3.16, 3.35] AC	2.33 [1.89, 2.76] CD	2.69 [2.55, 2.82] A	2.55 [2.46, 2.64] AC
Purple	CO04056-3P/PW	2.78 [2.68, 2.87] AC	3.03 [2.90, 3.15] AC	3.26 [3.14, 3.37] AC	2.52 [1.98, 3.07] AC	3.26 [3.11, 3.41] A	2.64 [2.50, 2.78] AC
	Purple Majesty	2.79 [2.65, 2.92] AB	3.09 [2.99, 3.19] AC	3.16 [3.05, 3.28] C	2.37 [1.99, 2.76] A	2.81 [2.76, 2.87] A	2.71 [2.63, 2.80] BC
Red	Crimson King	2.75 [2.61, 2.88] C	2.99 [2.90, 3.09] C	3.06 [2.96, 3.15] BC	2.23 [1.72, 2.74] BCD	2.76 [2.62, 2.90] A	2.51 [2.41, 2.62] AB
	Compound Code	*FU-15-1*	*FU-15-2*	*PA-15-1*	*PA-15-2*	*PZ-15*	*PR-15*
	Configuration	*Furan*		*Phenyl Acetate*		*Pyrazine*	*Pyrrole*
	Functional Group	*Ether*		*Ester*		*Nitrogenous*	

[a]Clone means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05) (n=10 x 14 clones).
[b]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.

Table 4.5. Log-transformed peak area cooking method analyte[a] means[b] with 95% confidence intervals detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014 and 2015

Year	*Compound Class*	Compound	*Compound Code*	Baked	Boiled
2014	*Aliphatic Aldehyde*	**2-phenylacetaldehyde**	*AD-14-5*	2.59 [2.52, 2.67]	2.43 [2.38, 2.48]
	Furan	**furfural**	*FU-14-2*	2.39 [2.31, 2.46]	2.18 [2.12, 2.23]
	Sulfur-Containing	**methional**	*SU-14*	2.41 [2.34, 2.49]	2.13 [2.05, 2.20]
2015	*Aliphatic Alkane*	**2,2,3,4-tetramethyl-pentane**	*HC-15-2*	3.40 [3.35, 3.45]	3.27 [3.22, 3.32]
		saturated hydrocarbon 1	*HC-15-3*	3.35 [3.31, 3.39]	3.29 [3.25, 3.32]
		saturated hydrocarbon 2	*HC-15-4*	3.36 [3.30, 3.41]	3.22 [3.16, 3.27]
		saturated hydrocarbon 3	*HC-15-5*	2.99 [2.95, 3.02]	2.88 [2.84, 2.92]
		unsaturated hydrocarbon 2	*HC-15-7*	3.21 [3.17, 3.25]	3.11 [3.08, 3.14]
	Cyclic Alkene	**azulene**	*HC-15-8*	3.28 [3.24, 3.31]	3.23 [3.20, 3.26]
	Aromatic Hydrocarbon	**3,4-dimethyl-styrene**	*HC-15-9*	2.42 [2.38, 2.46]	2.36 [2.34, 2.38]
	Terpene	**isomenthone**	*TE-15-3*	2.64 [2.58, 2.70]	2.47 [2.44, 2.50]
	Aliphatic Aldehyde	**(E)-2-heptenal**	*AD-15-2*	2.60 [2.54, 2.66]	2.51 [2.47, 2.54]
		pentanal	*AD-15-4*	3.65 [3.55, 3.76]	3.51 [3.45, 3.57]
	Aliphatic Ketone	**5-methyl-2-hexanone**	*KE-15-1*	2.99 [2.92, 3.06]	2.77 [2.74, 2.81]
	Aromatic Ketone	**p-methylaceto-phenone**	*KE-15-2*	2.97 [2.93, 3.01]	2.87 [2.84, 2.91]
	Cyclic Ketone	**3,4,5-trimethyl-2-cyclopenten-1-one**	*KE-15-3*	3.22 [3.13, 3.31]	3.01 [2.94, 3.07]
	Aliphatic Alcohol	**1-octen-3-ol**	*AC-15-1*	3.27 [3.23, 3.32]	3.12 [3.07, 3.17]
		1-pentanol	*AC-15-2*	3.32 [3.23, 3.41]	3.00 [2.95, 3.05]
		(Z)-2-methyl-2-penten-1-ol	*AC-15-3*	3.48 [3.37, 3.60]	3.32 [3.25, 3.40]
	Furan	**2-ethylfuran**	*FU-15-1*	3.31 [3.18, 3.43]	2.82 [2.78, 2.87]
		2-pentyl-furan	*FU-15-2*	3.20 [3.13, 3.28]	2.98 [2.94, 3.02]
		furfural	*FU-15-3*	3.41 [3.35, 3.46]	3.32 [3.28, 3.36]
	Halogen-Containing	**1-chloro-2-methyl-butane**	*HA-15*	3.18 [3.09, 3.27]	2.99 [2.93, 3.04]

[a]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.

[b]Cooking method means by analyte are significantly different according to ANOVA F-tests (α=0.05) and pairwise for cooking method by Welch's t-tests (α=0.05) (n=70-75 technical replicates x 2 cooking methods).

Table 4.6. Log-transformed peak area clone by cooking method means of analytes[a] with 95% confidence intervals detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014 and 2015

Flesh Color	Clone	Compound	Year	Baked	Boiled
White	**AC00395-2RU**	1-pentanol	2015	3.97 [3.80, 4.14]	3.03 [2.75, 3.31]
		2-ethylfuran	2015	4.15 [3.90, 4.41]	2.90 [2.81, 2.99]
		2-pentyl-furan	2015	3.67 [3.51, 3.84]	3.14 [3.01, 3.26]
		5-methyl-2-hexanone	2015	3.41 [3.30, 3.53]	2.75 [2.65, 2.86]
		1-chloro-2-methyl-butane	2015	3.75 [3.48, 4.03]	2.93 [2.76, 3.10]
	Fortress Russet	2-ethylfuran	2015	3.87 [3.63, 4.11]	2.82 [2.67, 2.96]
		1-chloro-2-methyl-butane	2015	3.36 [3.26, 3.45]	2.77 [2.51, 3.03]
		2-formyl-1-methylpyrrole	2015	2.65 [2.49, 2.82]	3.27 [3.13, 3.41]
	Russet Burbank	hexanal	2014	2.80 [2.62, 2.97]	3.70 [3.26, 4.14]
	Russet Norkotah	1-pentanol	2015	3.66 [3.47, 3.85]	2.95 [2.82, 3.08]
		2-ethylfuran	2015	3.81 [3.57, 4.05]	2.56 [2.47, 2.64]
	Russet Nugget	1-pentanol	2015	3.56 [3.51, 3.60]	2.90 [2.73, 3.07]
		(Z)-2-methyl-2-penten-1-ol	2015	4.92 [4.86, 4.99]	4.06 [3.91, 4.20]
		(E)-2-heptenal	2015	3.11 [3.05, 3.18]	2.57 [2.52, 2.61]
		pentanal	2015	4.44 [4.31, 4.57]	3.75 [3.64, 3.86]
		5-methyl-2-hexanone	2015	3.44 [3.20, 3.68]	2.75 [2.69, 2.81]
		3,4,5-trimethyl-2-cyclopenten-1-one	2015	3.97 [3.70, 4.25]	3.01 [2.90, 3.12]
		2-ethylfuran	2015	4.14 [4.00, 4.29]	3.07 [3.01, 3.12]
		2-pentyl-furan	2015	3.76 [3.58, 3.94]	3.08 [2.91, 3.25]
Yellow	**CO04067-8R/Y**	(Z)-2-methyl-2-penten-1-ol	2015	4.00 [3.82, 4.18]	3.10 [3.05, 3.15]
		benzaldehyde	2015	3.71 [3.67, 3.75]	3.17 [3.02, 3.32]
		pentanal	2015	3.93 [3.88, 3.98]	3.29 [3.23, 3.36]
	CO04099-3W/Y	5-methyl-2-hexanone	2015	3.41 [3.16, 3.66]	2.85 [2.65, 3.05]
		1-pentanol	2015	3.87 [3.50, 4.25]	3.14 [2.99, 3.29]
		2-ethylfuran	2015	3.33 [2.95, 3.71]	2.81 [2.58, 3.03]
	Masquerade	unidentifiable phenol	2015	3.46 [3.36, 3.55]	3.27 [3.15, 3.39]
		hexanal	2014	2.34 [2.08, 2.60]	3.18 [2.74, 3.63]
		(E)-2-heptenal	2015	3.12 [2.81, 3.43]	2.53 [2.48, 2.59]
		pentanal	2015	4.40 [4.32, 4.48]	3.78 [3.56, 4.01]
	Red Luna	hexanal	2014	2.57 [2.32, 2.83]	3.40 [2.76, 4.04]

[a]Clone by cooking method means by analyte are significantly different according to ANOVA F-tests (α=0.05) and pairwise for some clones by Tukey's HSD tests (α=0.05) (n=5 technical replicates x 2 cooking methods x 15 clones).

Table 4.7. Log-transformed peak area analyte[a] means and comparisons[b] with 95% confidence intervals by flesh color detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014

Flesh Color	n	(E,E)-2,4-nonadiene			3,5-dimethyl-cyclopentene		
White	48	1.88	[1.78, 1.98]	B	2.35	[2.22, 2.48]	C
Yellow	57	1.76	[1.68, 1.84]	AB	2.13	[2.02, 2.25]	B
Purple and Red	39	1.64	[1.59, 1.68]	A	1.87	[1.82, 1.92]	A
Compound Code		*HC-14-1*			*HC-14-2*		
Configuration		*Alkene*			*Cycoalkene*		
Functional Group		*Hydrocarbon*					

Flesh Color	n	(E)-2-heptenal			hexanal			2,4-nonadienal			(E)-2-octenal			2-phenyl-acetaldehyde		
White	48	2.51	[2.43, 2.6]	C	3.20	[3.05, 3.36]	C	2.25	[2.16, 2.34]	C	2.25	[2.16, 2.34]	C	2.51	[2.43, 2.59]	B
Yellow	57	2.29	[2.21, 2.37]	B	2.88	[2.72, 3.03]	B	2.11	[2.02, 2.19]	B	2.05	[1.96, 2.13]	B	2.62	[2.55, 2.69]	B
Purple and Red	39	2.11	[2.04, 2.18]	A	2.60	[2.5, 2.7]	A	1.92	[1.87, 1.97]	A	1.88	[1.82, 1.93]	A	2.34	[2.26, 2.42]	A
Compound Code		*AD-14-1*			*AD-14-2*			*AD-14-3*			*AD-14-4*			*AD-14-5*		
Configuration		*Aliphatic*														
Functional Group		*Aldehyde*														

Flesh Color	n	o-methyl-acetophenone			3,5-octadien-2-one			2-n-butyl furan		
White	48	1.99	[1.89, 2.08]	B	1.89	[1.79, 1.99]	B	1.94	[1.85, 2.03]	C
Yellow	57	1.90	[1.82, 1.98]	AB	1.79	[1.71, 1.88]	AB	1.77	[1.69, 1.86]	B
Purple and Red	39	1.79	[1.74, 1.85]	A	1.68	[1.63, 1.74]	A	1.63	[1.58, 1.68]	A
Compound Code		*KE-14-1*			*KE-14-2*			*FU-14-2*		
Configuration		*Aliphatic*						*Furan*		
Functional Group		*Ketone*						*Ether*		

[a]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.
[b]Flesh group means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05).

Table 4.8. Log-transformed peak area analyte[a] means and comparisons[b] with 95% confidence intervals by flesh color detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2015

Flesh Color	n	3-methyl-pentane	unsaturated hydrocarbon 1	unsaturated hydrocarbon 2	1-pentanol	(Z)-2-methyl-2-penten-1-ol
White	49	3.54 [3.45, 3.63] A	3.81 [3.73, 3.89] A	3.16 [3.13, 3.20] AB	3.29 [3.18, 3.40] B	3.59 [3.44, 3.74] B
Yellow	59	3.54 [3.46, 3.62] A	3.86 [3.80, 3.92] A	3.13 [3.08, 3.17] A	3.14 [3.06, 3.23] AB	3.37 [3.28, 3.45] A
Purple and Red	30	3.79 [3.70, 3.87] B	4.09 [4.02, 4.16] B	3.22 [3.17, 3.27] B	2.98 [2.91, 3.06] A	3.18 [3.10, 3.26] A
Compound Code		*HC-15-1*	*HC-15-6*	*HC-15-7*	*AC-15-2*	*AC-15-3*
Configuration		*Alkane*	*Alkene*		*Aliphatic*	
Functional Group		*Hydrocarbon*			*Alcohol*	

Flesh Color	n	benzaldehyde	(E)-2-heptenal	pentanal	5-methyl-2-hexanone	3,4,5-trimethyl-2-cyclopenten-1-one
White	49	3.45 [3.39, 3.51] B	2.61 [2.55, 2.67] B	3.66 [3.55, 3.76] B	2.97 [2.88, 3.06] B	3.26 [3.16, 3.37] B
Yellow	59	3.55 [3.49, 3.62] B	2.57 [2.52, 2.63] B	3.62 [3.51, 3.72] B	2.82 [2.76, 2.89] A	3.14 [3.06, 3.22] B
Purple and Red	30	3.28 [3.18, 3.37] A	2.43 [2.39, 2.46] A	3.38 [3.31, 3.46] A	2.87 [2.84, 2.91] AB	2.82 [2.74, 2.90] A
Compound Code		*AD-15-1*	*AD-15-2*	*AD-15-4*	*KE-15-1*	*KE-15-3*
Configuration		*Aliphatic*			*Aliphatic*	*Cyclic*
Functional Group		*Aldehyde*			*Ketone*	

Flesh Color	n	2-ethylfuran	2-pentyl-furan	2-isopropyl-3-methoxy-pyrazine	2-formyl-1-methylpyrrole
White	49	3.34 [3.17, 3.51] B	3.25 [3.16, 3.34] B	2.76 [2.70, 2.82] B	2.74 [2.67, 2.80] A
Yellow	59	2.99 [2.92, 3.06] A	2.99 [2.94, 3.04] A	2.72 [2.67, 2.77] A	2.62 [2.58, 2.67] A
Purple and Red	30	2.77 [2.70, 2.84] A	3.04 [2.98, 3.10] A	2.94 [2.84, 3.05] A	2.62 [2.55, 2.69] B
Compound Code		*FU-15-1*	*FU-15-2*	*PZ-15*	*PR-15*
Configuration		*Furan*		*Pyrazine*	*Pyrrole*
Functional Group		*Ether*		*Nitrogenous*	

[a]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.
[b]Flesh group means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05).

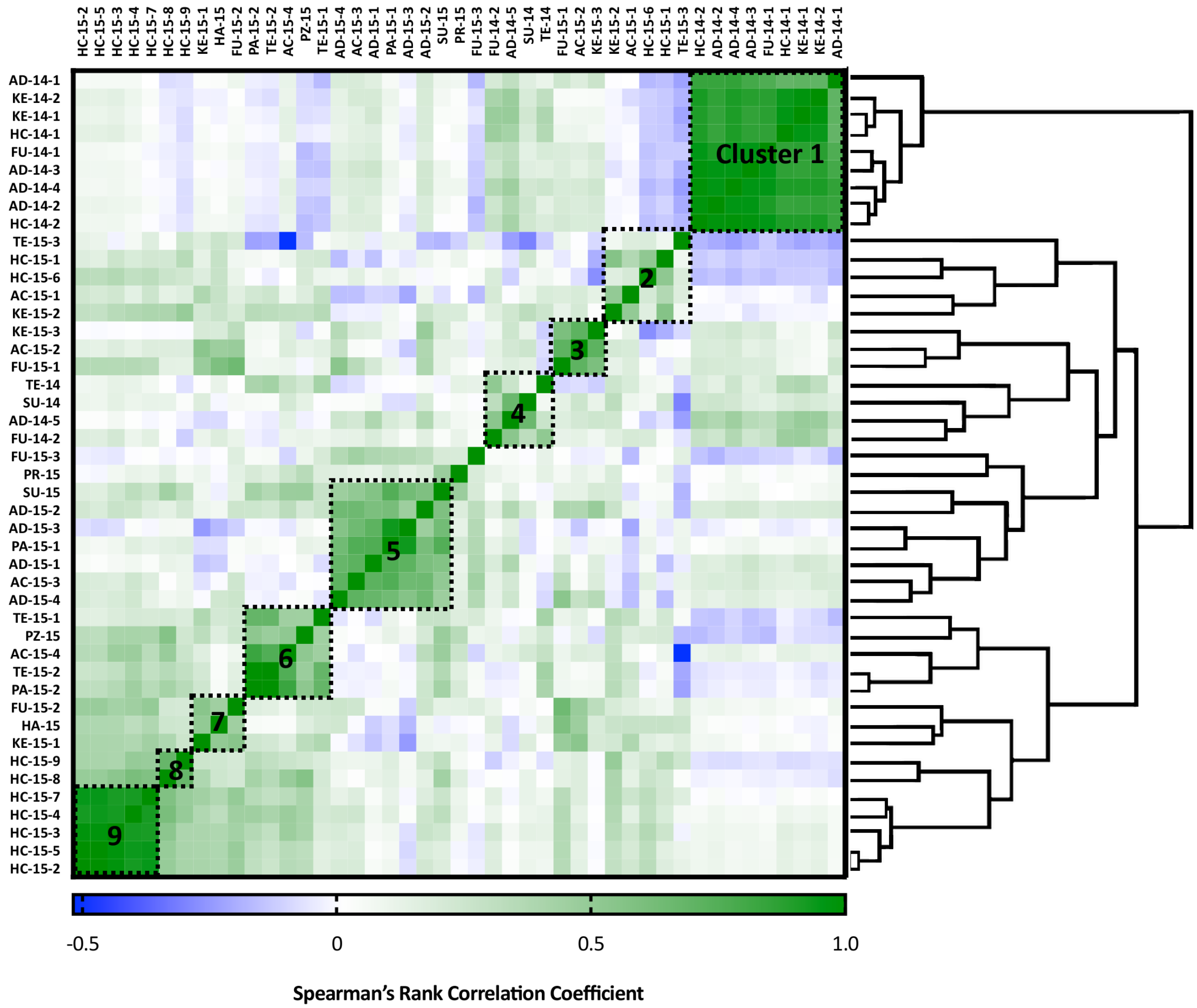

Figure 4.1. Spearman's rank correlation coefficients with two-way Ward HCA for analyte-analyte of cooked potato analytes using log-transformed peak areas (n=10 technical replicates x 15 clones) detected by non-targeted HS-SPME GC/MS

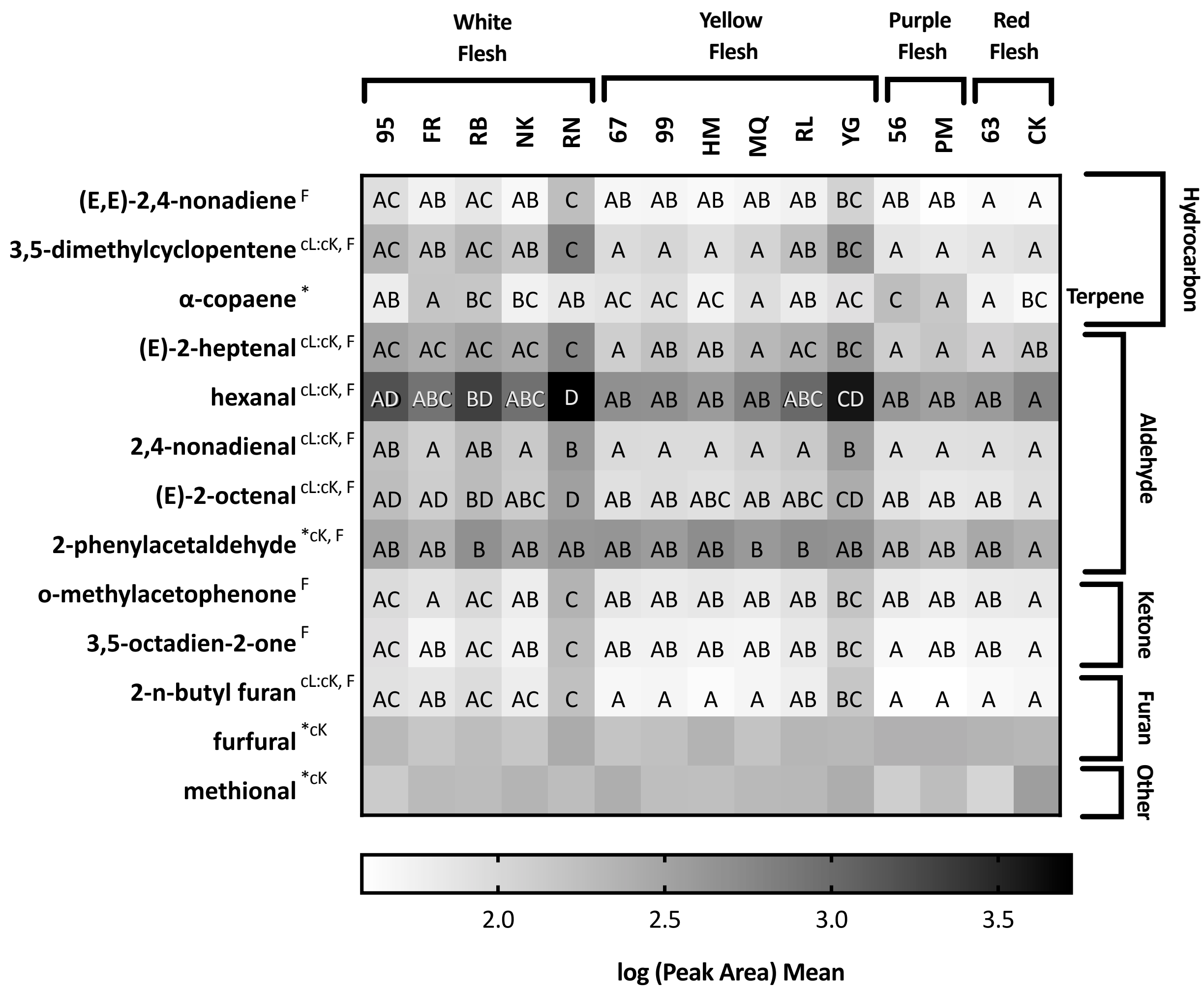

Fig. 4.2. Log-transformed peak area clone[a] means and comparisons[b] of analytes detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014

[a]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, 63=CO04063-4R/R, CK=Crimson King.

[b]Clone means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05) (n=technical replicates x 15 clones). Analytes are also significantly different according to mixed model F-tests (α=0.05) for cK=cooking method, cL:cK=clone by cooking method, or F=flesh color when notated.

*An asterisks indicates that analyte identification was confirmed using retention time matching of a standard in addition to spectral matching.

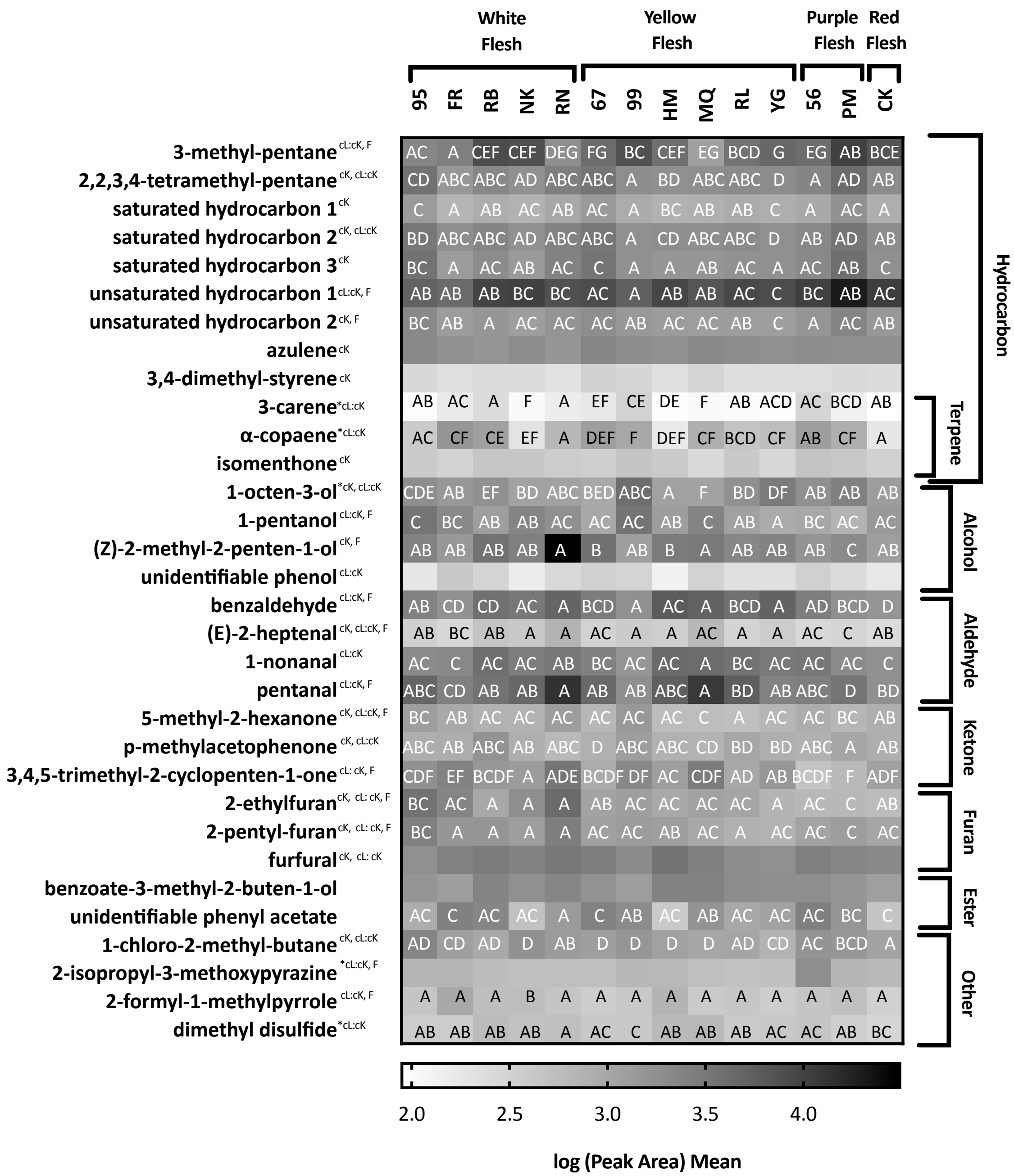

Figure 4.3. Log-transformed peak area clone[a] means and comparisons[b] of analytes detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2015

[a]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, CK=Crimson King.

[b]Clone means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05) (n=10 x 14 clones). Analytes are also significantly different according to mixed model F-tests (α=0.05) for cK=cooking method, cL:cK=clone by cooking method, or F=flesh color when notated.

*An asterisks indicates that analyte identification was confirmed using retention time matching of a standard in addition to spectral matching.

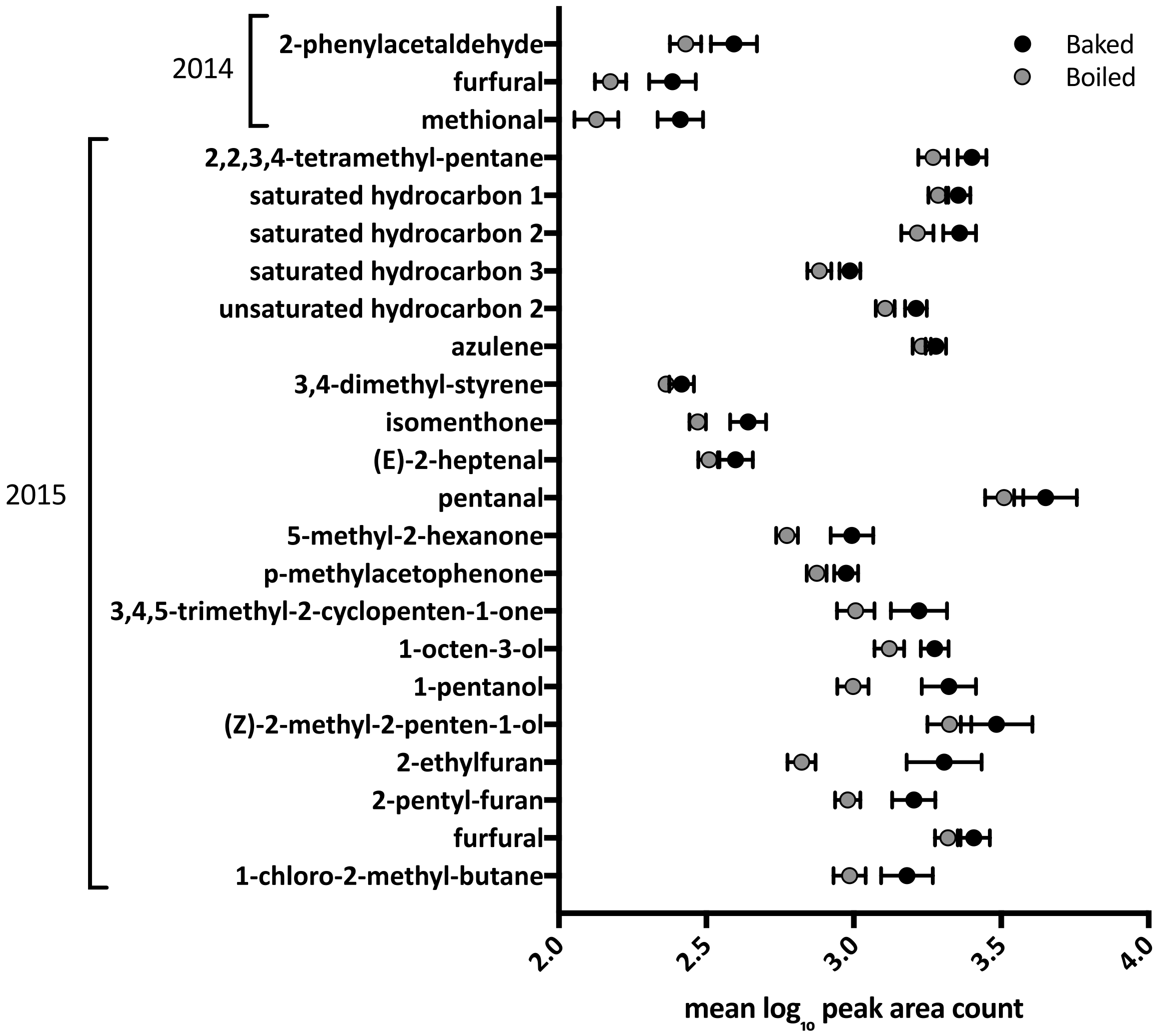

Figure 4.4. Log-transformed peak area cooking method means of analytes[a] with 95% confidence intervals detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014 and 2015

[a]Cooking method means by analyte are significantly different according to ANOVA F-tests (α=0.05) and pairwise for cooking method by Welch's t-tests (α=0.05) (n=70-75 technical replicates x 2 cooking methods).

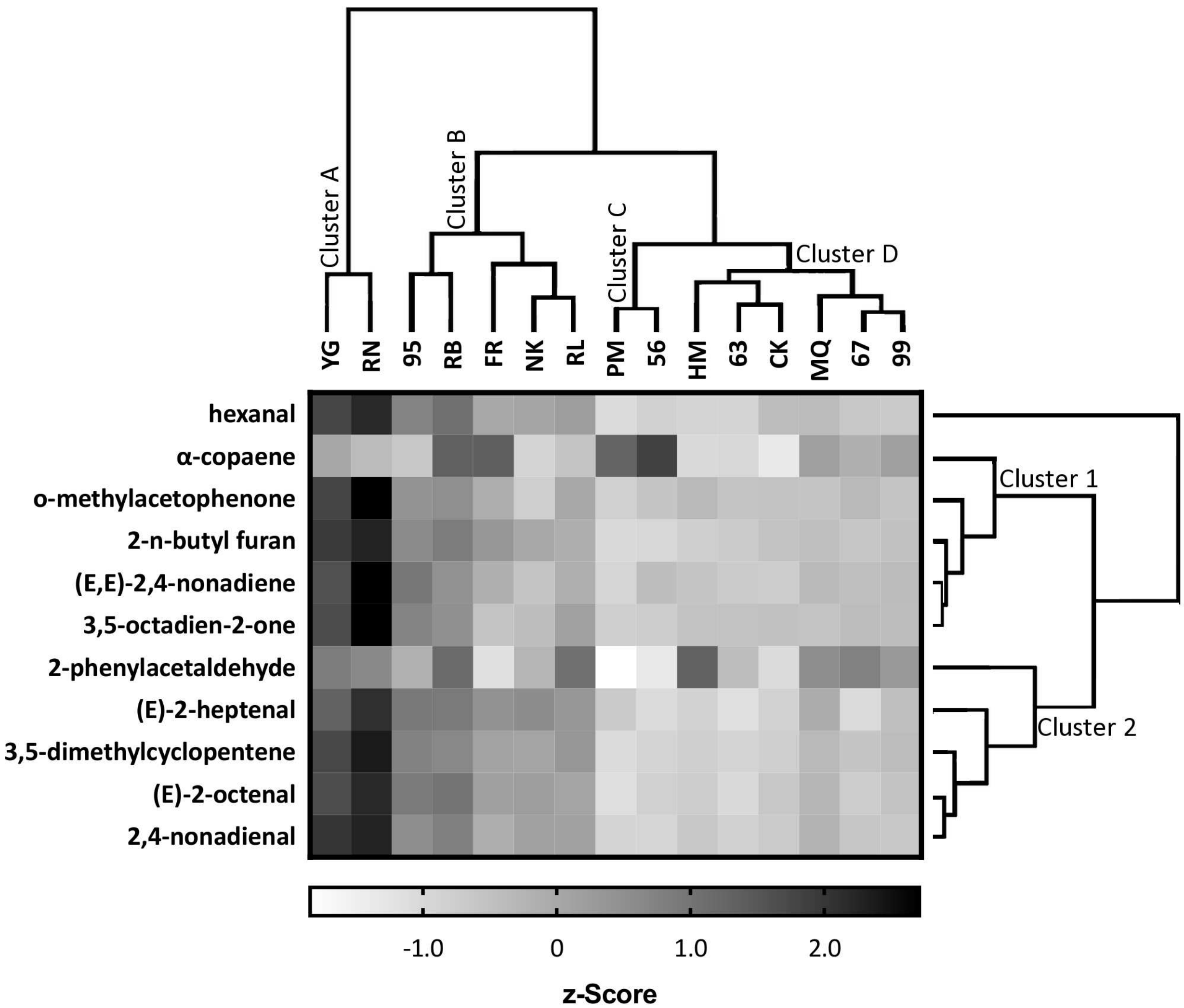

Figure 4.5. Log-transformed peak area clone[a] means of analytes[b] with z-score normalization and two-way Ward HCA, detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014

[a]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, 63=CO04063-4R/R, CK=Crimson King.

[b]Clone means by analyte are significantly different according to ANOVA F-tests (α=0.05) (n=10 technical replicates x 15 clones).

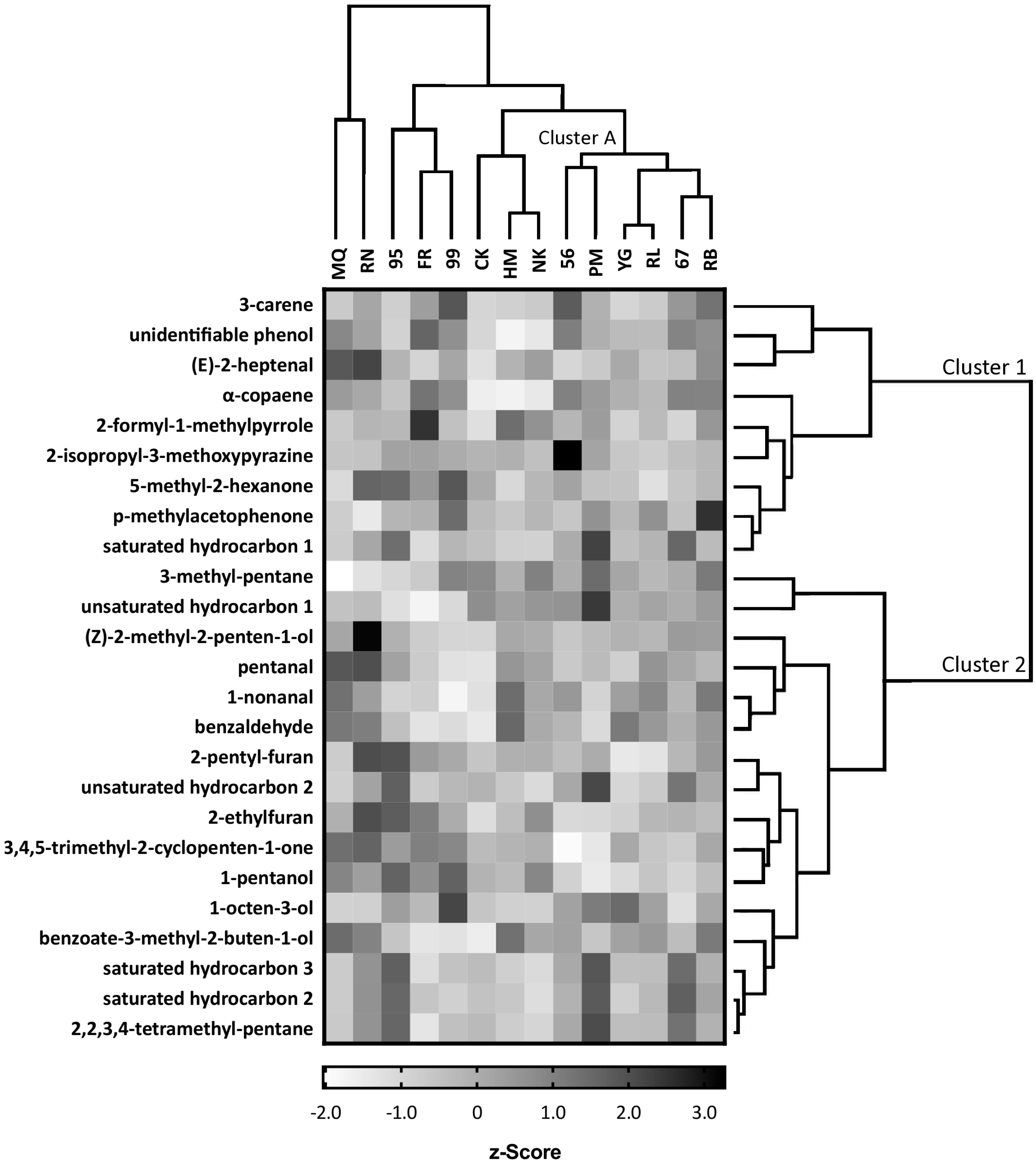

Figure 4.6. Log-transformed peak area clone[a] means of analytes[b] with z-score normalization and two-way Ward HCA, detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2015

[a]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, CK=Crimson King. .

[b]Clone means by analyte are significantly different according to ANOVA F-tests (α=0.05) (n=10 technical replicates x 14 clones).

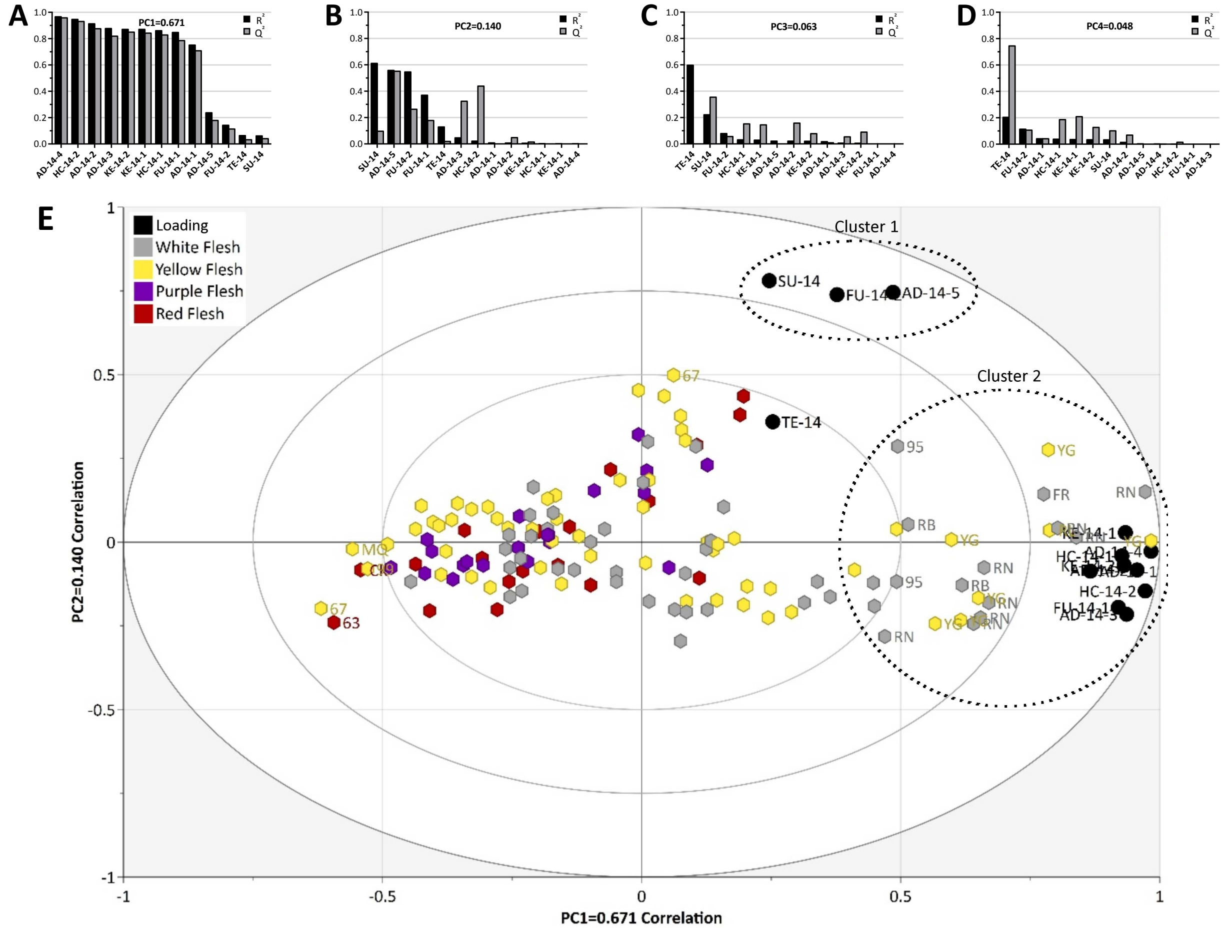

Figure 4.7. (A-D) Loading contributions and (E) normalized correlation R^2 biplot in principle component analysis of Pareto-scaled analyte[a,b] loadings and potato clone[c] by cooking method scores colored by flesh color[d], analyte detection by HS-SPME GC/MS in 2014

[a]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.

[b]All analytes are significantly different according to ANOVA F-tests (α=0.05) for clone, cooking method, or clone and cooking method interactions (n=5 technical replicates x 2 cooking methods x 15 clones).

[c]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, 63=CO04063-4R/R, CK=Crimson King.

[d]Only scores with $R^2 \geq 0.50$ are labeled with clone codes.

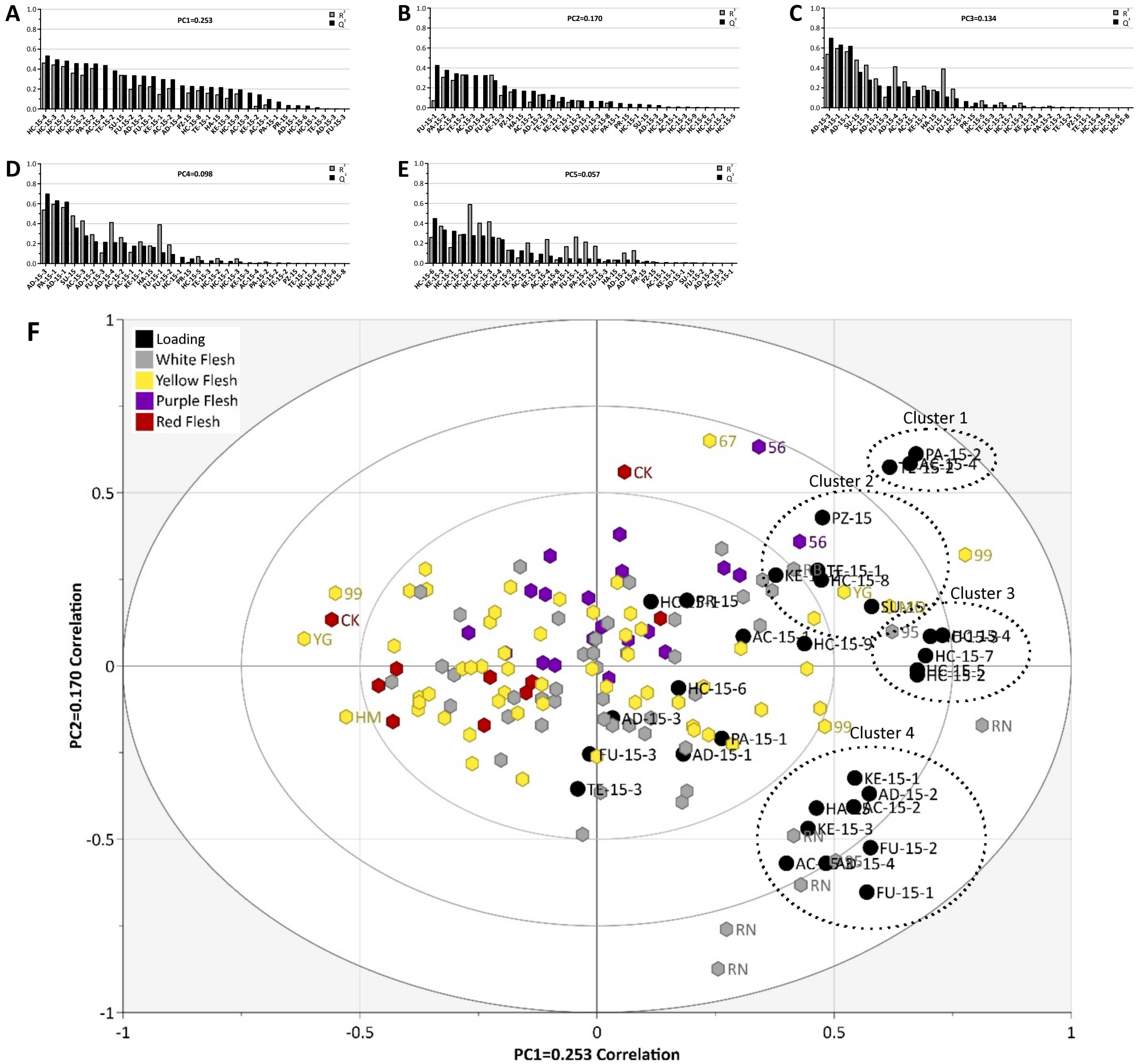

Figure 4.8. (A-E) loading contributions and (F) normalized correlation R^2 biplot in principle component analysis of Pareto-scaled analyte[a,b] loadings and potato clone[c] by cooking method scores colored by flesh color[d], analyte detection by HS-SPME GC/MS in 2015

[a]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.

[b]All analytes are significantly different according to ANOVA F-tests (α=0.05) for clone, cooking method, or clone and cooking method interactions (n=5 technical replicates x 2 cooking methods x 14 clones).

[c]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, CK=Crimson King.

[d]Only scores with R^2≥0.50 are labeled with clone codes.

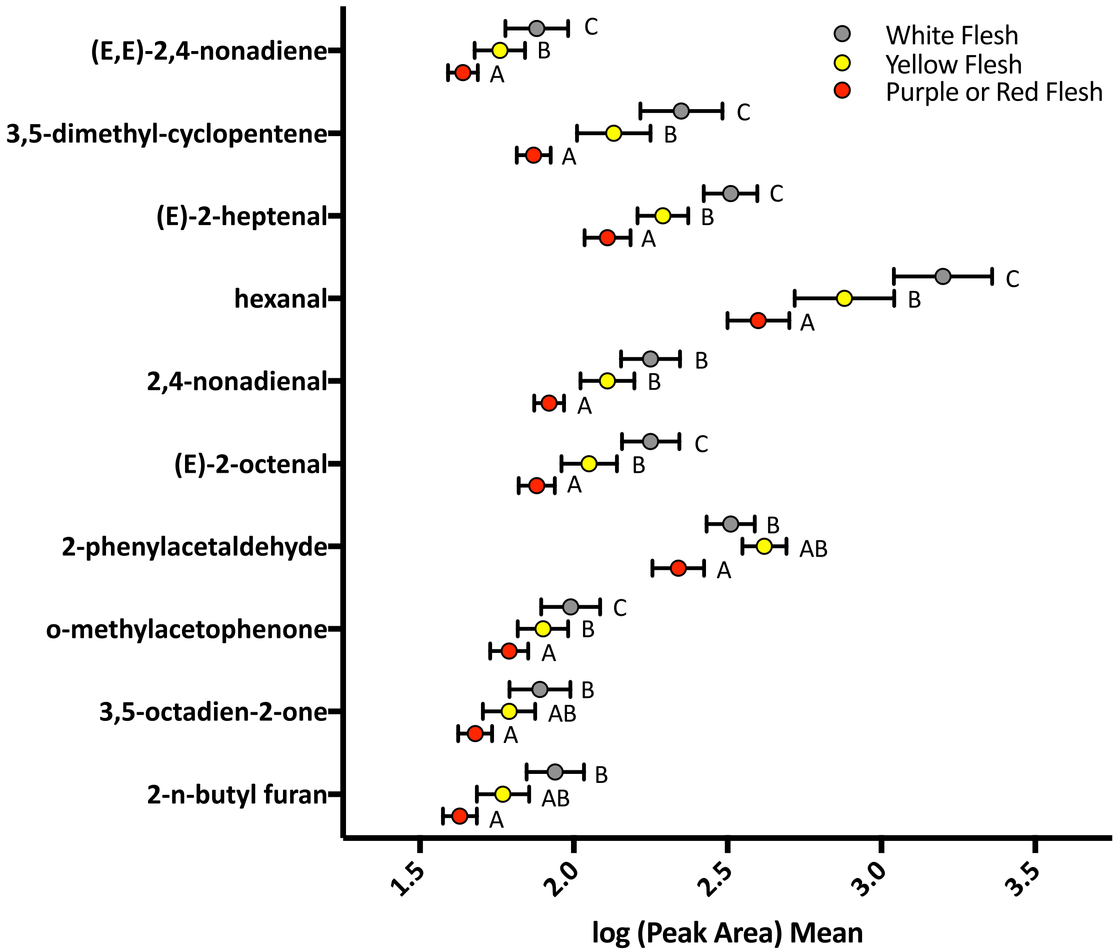

Figure 4.9. Log-transformed peak area means[a] and comparisons[b] of analytes with 95% confidence intervals by flesh color detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2014

[a]Flesh group clone number: n=5 for white, n=6 for yellow, n=4 for red.

[b]Flesh group means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05).

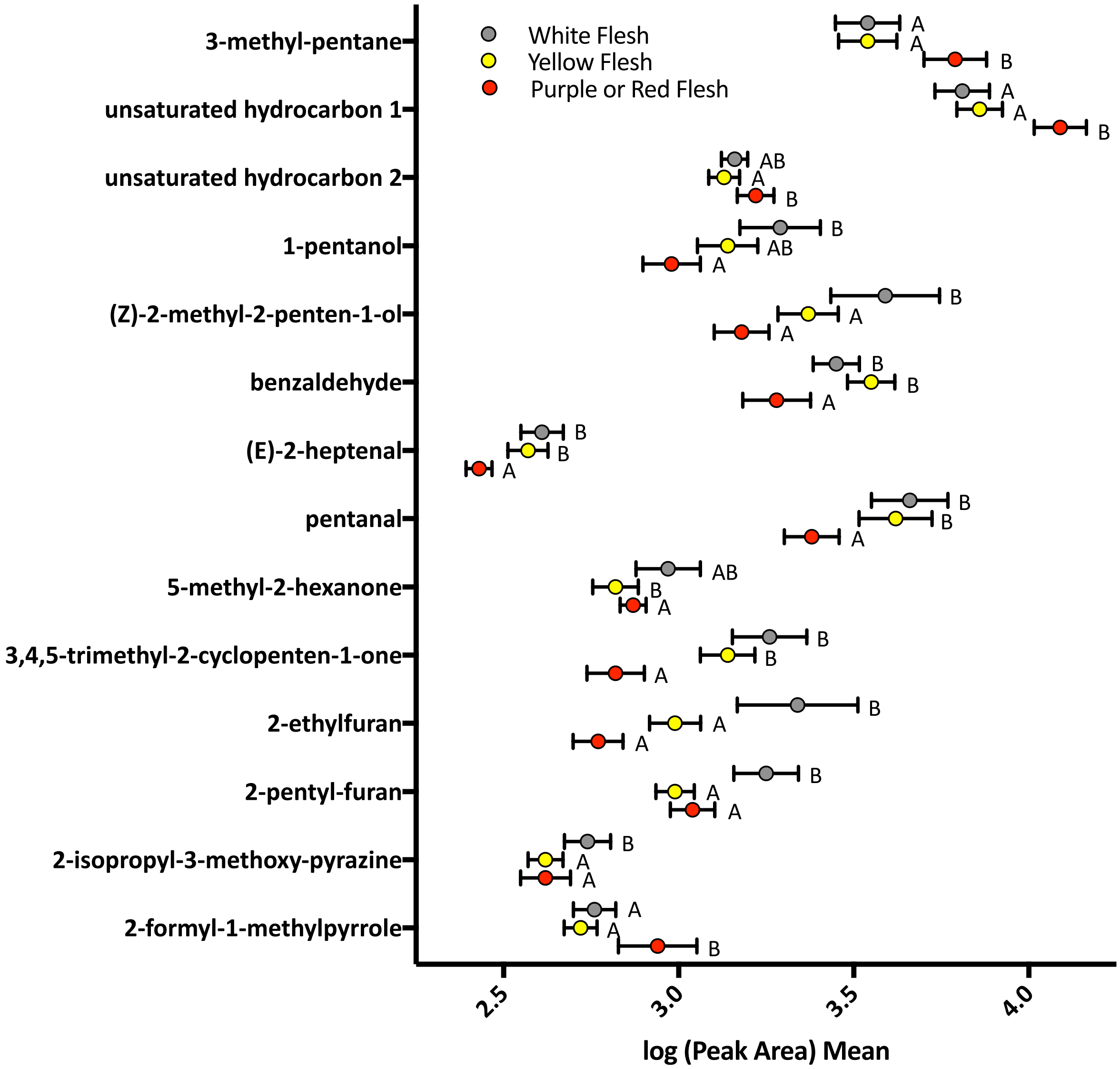

Figure 4.10. Log-transformed peak area means[a] and comparisons[b] of analytes with 95% confidence intervals by flesh color detected by non-targeted HS-SPME GC/MS in cooked potatoes in 2015

[a]Flesh group clone number: n=5 for white, n=6 for yellow, n=3 for red.

[b]Flesh group means by analyte are significantly different according to ANOVA F-tests (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Tukey's HSD test (α=0.05).

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CHAPTER 5. COMBINED SENSORY AND INSTRUMENTAL TEXTURE ANALYSES

5.1. Introduction

Texture influences flavor taste and aroma compound release through density, cohesion, adhesion, malleability, and other factors.[1] In potatoes, starch content, related to specific gravity, is a fairly reliable indicator of sensory texture, particularly in terms of mealiness versus waxiness.[2] Russets and other white-fleshed potatoes tend to have a greater starch content than yellow-, purple-, or red-fleshed potatoes, therefore resulting in a grainy, dry, mealy texture in contrast to waxiness, which is gummier and smoother.[3] At least one recent sensory study of potato demonstrates sensory texture differentiation among clones.[4] In this study, sensory scores were used to evaluate objective instrumental measurements for hardness of cooked potatoes. Clones and cooking methods were compared.

5.2. Materials and Methods

5.2.1. Preparation

Potato tubers included advanced breeding selections (number codes) and named cultivars with a range of skin types and flesh colors. White-fleshed clones were AC00395-2RU, Fortress Russet, Russet Burbank, Russet Norkotah, and Russet Nugget; yellow-fleshed clones were CO04067-8R/Y, CO04099-3W/Y, Harvest Moon, Masquerade, Red Luna, and Yukon Gold; purple-fleshed clones were CO04056-3P/PW and Purple Majesty; red-fleshed clones were CO04063-4R/R and Crimson King. Clones were field grown and conditioned at the Colorado State University San Luis Valley Research Center (SLVRC, Center, CO) in 2014 and 2015. Whole tubers were baked in aluminum foil or boiled in water, diced, and served warm to a sensory panel or kept warm until instrumental texture analysis.

5.2.2. Sensory Analysis

A trained sensory panel of 15 volunteers conducted a descriptive flavor analysis of potatoes cooked by both methods from both growing seasons. Panelists were 47% men and 53% women; 80% white, 20% other races; 20% aged 18 to 25, 27% aged 26 to 40, 27% aged 41 to 55, and 27% aged 56 to 70. Training included a discussion of potato flavor, aroma, appearance, and texture. By consensus, a scorecard was developed for 15 attributes with a number-anchored hedonic scale from 1 to 9, absence to maximum intensity. Panelists scored each clone by cooking method twice over both years. Individual diced potato samples contained material from at least three tubers.

5.2.3. Instrumental Texture Analysis

A CT3-10kg Texture Analyzer (Brookfield Engineering, Middleboro, MA) was used for instrumental texture analysis. Tubers were halved longitudinally. Skin was removed from one half and load measurements were taken with a spherical probe for both halves, flesh side down. In 2014, analysis included 3 to 9 tubers per clone by cooking method, whereas 2015 analysis evaluated 30 tubers per clone by cooking method. Maximum loads, representative of hardness, were compiled from each tuber half.

5.2.4. Statistical Analysis

Mealy texture score clone mean estimates were calculated by residual maximum likelihood (REML) in a linear mixed model in Rstudio version 0.99.896 (RStudio, Inc., Boston, MA) with clone, cooking method, and their interaction as fixed effects and panelist, order, year and interactions as random effects. A similar linear mixed model was used for maximum load (hardness) mean estimates from instrumental measurements, but with only year and interactions with fixed effects as random effects. Only effects significant by a mixed model F-test (α=0.05) were used for least square mean estimates and Satterthwaite t-test contrasts (α=0.05). Regression of clone sensory mean estimates versus clone maximum load mean estimates was assessed.

5.3. Results and Discussion

Clone is a significant effect in the sensory mealy texture mixed model (Figure 5.1; Table S3.2), but not the instrumental mixed model. Cooking method is significant for both models. Additional significant effects in the instrumental model are the interactions of clone with cooking method and clone with year. Panelist and the interaction of panelist with cooking method are significant effects in the sensory model. Flesh color is not significant in a mixed model with flesh color rather than clone for sensory mealy texture or instrumental hardness.

Russet Norkotah and Fortress Russet maximum load mean estimates are greater than most other clones, though this trend is not significant (Table 5.1; Figure 5.2). The sensory mealy texture score mean estimate for baked potatoes is greater than the boiled potato score mean estimate. Conversely, load mean estimates are greater for boiled versus baked potato totals for overall baked mean estimates (Table 5.2), which is reflected by clone and cooking method interaction mean estimates for eleven clones (Table 5.3; Figure 5.3). Instrumental max loads and sensory scores by clone do not correlate linearly or non-linearly in regression.

Although both instrumental hardness measurements and sensory mealiness scores have significant variation by clone, the variables are seemingly unrelated when clone means are used as the sample population. Disparity between instrumental and texture measurements is not uncommon,[1] but may suggest inconsistencies in one or both measurements. For sensory analysis, food references were not provided for mealy texture, which may have resulted in poor distinction by panelists. Uneven cooking may have also played a role. Instrumental analysis was relatively standardized, but texture can change as potatoes cool. Measurements were not randomized by clone, so some clones may have experienced cooling. Still, both separate analyses distinguish between baked or boiled potatoes, where baked potatoes have a greater sensory mealy score mean estimate and smaller maximum mean load estimate compared to boiled potatoes. Baking tends to dry potato flesh compared to boiling, which explains greater mealy sensory scores but smaller hardness measurements for baked potatoes. Boiled potatoes retain more water, which would make mouthfeel gummier rather than mealy, resulting in lower mealy score ratings. The retention of water also means more turgid cells, and therefore, greater hardness.

5.4. Summary

In this study, the ability of an instrumental texture analyzer to measure mealiness of cooked potatoes was assessed by evaluating the relationship between sensory texture scores for mealiness and instrumental texture measurements for hardness. Trained sensory panel ratings were used to calculate mealy texture score mean estimates by clone, which had some significant differences (linear mixed model ANOVA F-test and Satterthwaite t-tests, $\alpha=0.05$, n=17-38 ratings per clone). Instrumental texture hardness mean estimates were not significantly different by clone. Baked potato sensory mealy scores were significantly greater than boiled potato scores, whereas baked potato hardness measurements were significantly less than that of boiled potatoes. Instrumental hardness measurements and sensory scores were not correlated in regression of variable means by clone.

Table 5.1. Max load (hardness) clone mean estimates[a,b] with 95% confidence intervals for cooked potatoes

Flesh Color	Clone	Max Load (g)		
White	**AC00395-2RU**	985	[717, 1253]	EF
	Fortress Russet	1308	[1035, 1582]	BCDE
	Russet Burbank	945	[681, 1210]	ABCDE
	Russet Norkotah	1309	[1043, 1576]	ABCD
	Russet Nugget	933	[667, 1199]	G
Yellow	**CO04067-8R/Y**	823	[554, 1092]	EF
	CO04099-3W/Y	879	[611, 1146]	CDEF
	Harvest Moon	935	[662, 1208]	DEF
	Masquerade	834	[565, 1102]	ABC
	Red Luna	926	[657, 1195]	AB
	Yukon Gold	976	[708, 1244]	DEF
Purple	**CO04056-3P/PW**	853	[585, 1121]	A
	Purple Majesty	694	[420, 967]	FG
Red	**CO04063-4R/R**	717	[330, 1103]	ABCDEF
	Crimson King	1034	[769, 1299]	DEF

[a]Means estimated using REML in a mixed model analysis with year as a random effect and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=33-39 tuber replicates per clone).

[b]Clone means are not significantly different according to a mixed model F-test (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Satterthwaite t-tests (α=0.05).

Table 5.2. Max load (hardness)[a] and hedonic score (1 to 9, absence to maximum intensity)[b] mealy texture cooking method mean estimates with 95% confidence intervals for cooked potatoes

	Baked		Boiled	
Max Load (g)	821	[746, 896]	1066	[992, 1140]
Sensory	4.01	[3.27, 4.75]	3.10	[2.34, 3.87]

[a]Max load means estimated using REML in a mixed model analysis with year as a random effect and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=33-39 tubers per clone); Means are significantly different according to a mixed model F-test (α=0.05) and pairwise comparison of means in Satterthwaite t-tests (α=0.05)

[b]Sensory mealy texture means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=237-291 tuber replicates x 2 cooking methods); Means are significantly different according to a mixed model F-test (α=0.05) and pairwise comparison of means in Satterthwaite t-tests (α=0.05).

Table 5.3. Max load (hardness) clone by cooking method mean estimates[a,b] with 95% confidence intervals for cooked potatoes

Flesh Color	Clone	Baked		Boiled		
White	**AC00395-2RU**	860	[582, 1138]	1110	[835, 1385]	
	Fortress Russet	1128	[844, 1413]	1489	[1208, 1769]	
	Russet Burbank	849	[575, 1122]	1042	[771, 1313]	
	Russet Norkotah	1355	[1076, 1633]	1264	[993, 1535]	ns
	Russet Nugget	796	[522, 1071]	1070	[797, 1343]	
Yellow	**CO04067-8R/Y**	636	[356, 915]	1010	[735, 1285]	
	CO04099-3W/Y	917	[642, 1192]	840	[564, 1116]	ns
	Harvest Moon	670	[388, 951]	1200	[920, 1481]	
	Masquerade	797	[514, 1079]	871	[599, 1143]	ns
	Red Luna	619	[338, 899]	1234	[959, 1508]	
	Yukon Gold	816	[539, 1093]	1136	[861, 1411]	
Purple	**CO04056-3P/PW**	757	[481, 1033]	949	[672, 1225]	
	Purple Majesty	587	[304, 870]	800	[518, 1082]	ns
Red	**CO04063-4R/R**	733	[307, 1158]	700	[294, 1107]	ns
	Crimson King	794	[520, 1068]	1274	[1002, 1546]	

[a]Means estimated using REML in a mixed model analysis with year as a random effect and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=33-39 tuber replicates per clone).

[b]Clone by cooking interaction is significant according to a mixed model F-test (α=0.05) and pairwise for some clones by Satterthwaite t-tests (α=0.05), unless noted with ns=not significant.

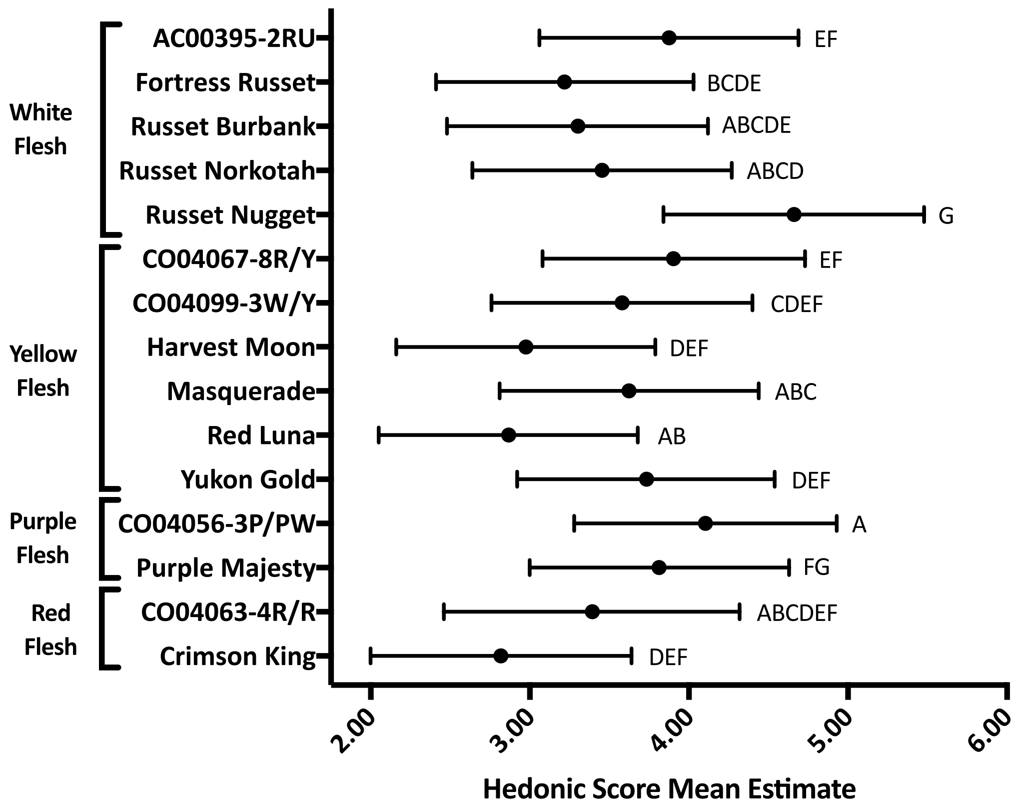

Figure 5.1. Hedonic score (1 to 9, absence to maximum intensity) mealy texture clone mean estimates[a] and comparisons[b] with 95% confidence intervals of cooked potato sensory attributes rated by a trained sensory panel

[a]Means estimated using REML in a mixed model analysis with year as a random effect and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=33-39 tuber replicates per clone).

[b]Clone means are significantly different according to a mixed model F-test (α=0.05), where means labeled with the same capital letter are not significantly different but different capital letters indicate significantly different means in Satterthwaite t-tests (α=0.05).

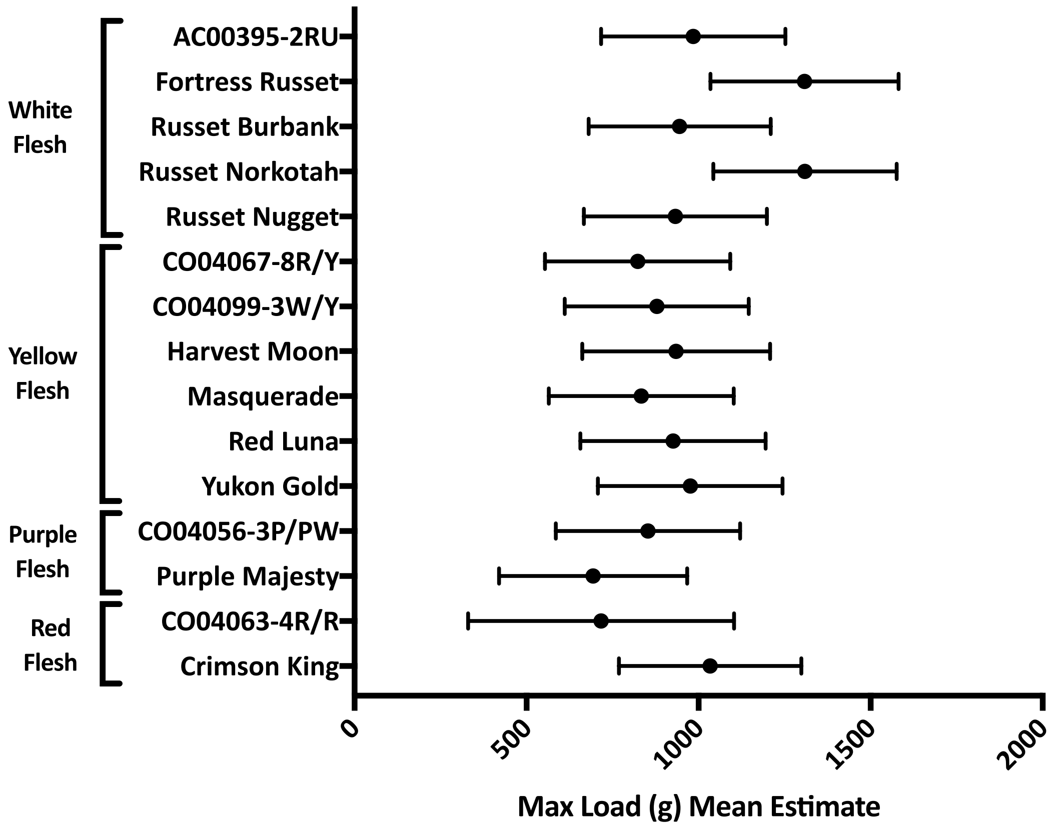

Figure 5.2. Max load (hardness) clone mean estimates[a,b] with 95% confidence intervals for cooked potatoes

[a]Means estimated using REML in a mixed model analysis with year as a random effect and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=33-39 tuber replicates per clone).

[b]Clone means are not significantly different according to a mixed model F-test (α=0.05).

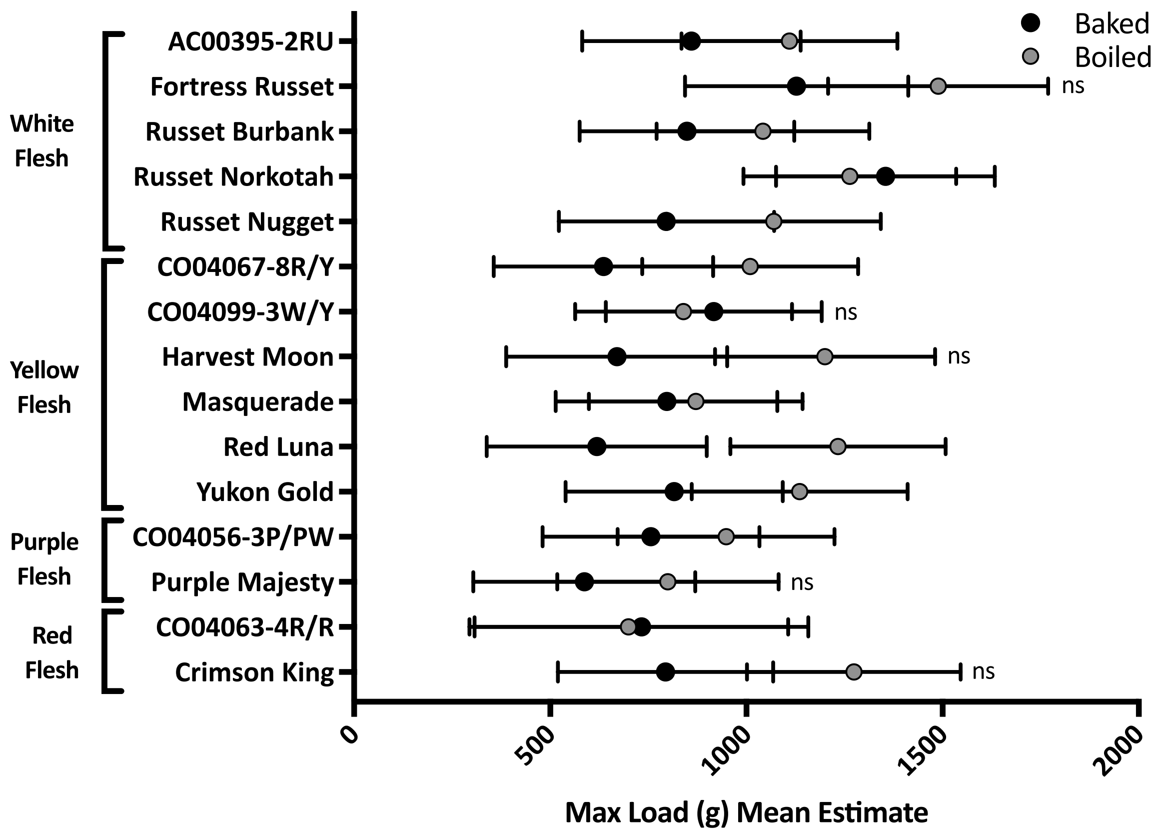

Figure 5.3. Max load (hardness) clone by cooking method mean estimates[a,b] with 95% confidence intervals for cooked potatoes

[a]Means estimated using REML in a mixed model analysis with year as a random effect and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=33-39 tuber replicates per clone).

[b]Clone by cooking interaction is significant according to a mixed model F-test (α=0.05) and pairwise for some clones by Satterthwaite t-tests (α=0.05), unless noted with ns=not significant.

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CHAPTER 6. COMBINED SENSORY AND AROMA COMPOUND ANALYSES

6.1. Introduction

Flavor is a subjective, complex quality trait conferred by non-volatile, semi-volatile, or volatile compounds.[1,2] Perception of flavor may be influenced by appearance, texture, personal preference, or other characteristics.[3] Taste is the basic reception of typically non-volatile metabolites that produce bitter, sweet, salty, sour, and umami sensations on the tongue.[4] In potatoes, umami and bitter are most prominent, whereas the other tastes are minimal.[1] Aroma is the reception of volatile or semi-volatile compounds in the olfactory that produce more complex sensations and greater contributions to flavor compared to taste,[1,3] even at small concentrations.[2]

Most potato aroma compounds result from thermal cooking interactions between fatty acid, sugar, and amino precursors, particularly aldehyde formation through Strecker amino acid degradation, aldehyde and ketone formation via lipid degradation, pyrazine formation in Maillard reactions, and volatile products from other sugar degradation reactions.[5,6] Prominent volatile flavor compounds extant in raw tubers include sulfur metabolites, terpenes, and pyrazines.[6–8] Several comprehensive aroma profiles exist for fried, chipped, baked, microwaved, boiled, or dehydrated potatoes. Potato aroma profiles have been found to differ by cultivar,[9–11] environment,[10] storage conditions,[6,12] and cooking method.[11]

Though past aroma compound extraction methods are slow and resource-intensive, headspace solid phase-microextraction (HS-SPME) provides a fast, solvent-free method that enables more efficient aroma phenotyping[13,14] when used in combination with gas chromatography (GC) and mass spectrometry (MS). A handful of volatile profiles for cooked European potato cultivars have been generated through HS-SPME GC-MS,[15–18] but minimal information exists for cooked American potato cultivars using this method. Moreover, very few volatile assays have been paired to sensory analysis to confirm flavor compounds. Unlike other potato flavor studies with a sensory component, the following sensory analysis covers an extensive array of flavor attributes for identification of aroma compounds significant to cooked fresh market potato flavor. Confirmed aroma compounds provide a framework for the development of flavor biomarker assays and gene discovery studies. Further development of potato aroma compound detection methods may provide a means of high-throughput flavor phenotyping using select biomarkers, which would be particularly useful in selection-based breeding programs for flavor improvement.

6.2. Materials and Methods

6.2.1. Tuber Material and Preparation

The Colorado State University (CSU) Potato Breeding and Selection program provided potato tubers field grown at the CSU San Luis Valley Research Center (SLVRC, Center, CO). During 2014 and 2015 growing seasons, potatoes received conventional fertilizer, pesticide, and drip-irrigation applications. Tuber material comprised advanced selections and named cultivars developed by the CSU Potato Breeding and Selection program, including white-fleshed clones AC00395-2RU, Fortress Russet, and Russet Nugget; yellow-fleshed clones CO04067-8R/Y, CO04099-3W/Y, Harvest Moon, Masquerade, and Red Luna; purple-fleshed clones CO04056-3P/PW and Purple Majesty; and red-fleshed clones CO04063-4R/R and Crimson King. Additional commercial cultivars were white-fleshed Russet Burbank and Russet Norkotah clones, as well as yellow-fleshed clone Yukon Gold. Storage-conditioned potato tubers for each clone were baked or boiled, diced into cubes, and served warm to sensory panelists or frozen with liquid N_2 for storage at -80°C until volatile analysis. Bulked, randomized samples for each clone by cooking method contained material from at least three tubers per individual sample in both sensory and volatile analyses. In sensory analysis, presented sample codes and order were random.

6.2.2. Sensory Analysis

A trained sensory panel evaluated each potato clone by cooking method once per growing season, with at least eight panelists present in a sensory session. Fifteen panelists were recruited as volunteers in the San Luis Valley of Colorado. Demographics were 47% men and 53% women; 80% white, 20% other races; 20% aged 18 to 25, 27% aged 26 to 40, 27% aged 41 to 55, and 27% aged 56 to 70. Training included potato flavor discussions, aroma sampling, description of active flavor enhancement while eating, reference food consensus, and scorecard development. Using a categorical scale from 1 to 9 with both numeric and hedonic anchors (from 0 or absence to 9 or maximum attribute intensity), the following sensory attributes were rated: appearance, aroma intensity, potato-like flavor, sweet, fruity, lemon, umami, buttery, creamy, earthy, woody, bitter, off-flavors other than bitter, mealy texture, and overall quality (see scorecard in Appendix 1). Panelists received reference foods, water, and crackers in each sensory session. Samples were presented with three-digit codes. To account for order effects, sample order was randomized across panelists.

6.2.3. Aroma Compound Analysis

A non-targeted metabolomics workflow was used for cooked potato aroma compound analysis. For each clone by cooking method, 5.0 g of frozen, cooked potato was transferred to 20 mL HS vials (75.5 x 22.5 mm; MicroSolv Technology Co., Leland, NC), gently thawed, mashed, and resealed. Vial temperatures were equilibrated by immersion in an 80°C water bath for 5 minutes prior to volatile extraction. For volatile adsorption, a 75 µm carboxen-coated 23 mm polydimethylsiloxane (CAR/PDMS) SPME fiber (Supelco Inc., Bellefonte, PA) was exposed to vial HS for 20 minutes at 50 °C and 300 rpm, followed by desorption and splitless injection with helium carrier gas at a constant flow rate of 1.5 mL min^{-1} for 2 minutes at 280°C. An Agilent 7890A (Agilent Technologies, Santa Clara, CA) equipped with an Agilent VF5-ms (5% phenyl-methyl) capillary GC column separated analytes using temperature ramps from 35 to 280°C. After internal electron ionization at 70 eV, an Agilent 240 ion trap was used to detect analyte fragments in full scan mode from 29-400 m/z at 0.45 seconds per scan. The HS-SPME GC-MS method was automated with System Control in MS Workstation version 6 (Agilent Technologies). Analysis consisted of five technical replications in a randomized order.

Mass spectrometry data was processed with the XCMS package in RStudio version 0.99.896 (RStudio, Inc., Boston, MA). Peaks were detected using a signal to noise ration (S/N) of 2, subsequently corrected for retention time, and finally normalized using the total ion current (TIC). Peaks were clustered and correlated by mass abundance and retention time with the package 'ramclustR' in Rstudio, generating reconstructed mass spectra for analyte identification. To focus on aroma compounds contributing to variation across samples, an analysis of variance (ANOVA) with false discovery rate adjustments and principal component analysis (PCA) with Pareto-scaling was used to eliminate analytes without significant variation (F-test, α=0.05). Analytes with significant variation were identified using spectral matching of reconstructed spectra in National Institute of Standards and Technology (NIST) MS Search Software version 2.0f (NIST, Gaithersburg, MD) with the NIST 11 database and a minimum intensity matching threshold of 450. Retention time matching with a user generated database of standards supplemented spectral matching for 3-carene, α-copaene, dimethyl disulfide, furfural, methional, 2-isopropyl-3-methoxypyrazine, 1-octen-3-ol, and 2-phenylacetaldehyde.

6.2.4. Statistical Analysis

To account for unbalanced sample numbers, univariate linear mixed models were used to identify sensory attributes with significant variation in fixed effects (ANOVA, F-test, α=0.05). Fixed effects were clone, and the interaction between clone and cooking method (n= 8-21 hedonic scores x 15 clones x 2 cooking methods) while random effects were session year, sample order presented to panelists, panelists, and interactions. Clone, cooking method, and clone by cooking method interaction attribute means were estimated using residual maximum likelihood (REML) least square means. Variable sensory attributes by clone were confirmed by linear least square mean contrasts with Welch's adjustments (t-test, α=0.05). Sensory attribute linear mixed models and statistical tests were calculated with the 'lme4' package in Rstudio version 0.99.896 (RStudio, Inc., Boston, MA).

Mean peak areas of aroma compounds by clone, cooking method, and the interaction between clone and cooking method were calculated (n=10 technical replications). Aroma compounds with significant variation between mean peak areas by clone according to ANOVA F-tests (α=0.05) and Tukey's Honest Significant Difference (HSD) multiple comparison tests (t-tests, α=0.05) were used for further analysis.

Sensory score mean estimates and aroma compound peak area means by clone were used for multivariate analysis. Spearman's rank correlation coefficients were calculated between sensory attributes and aroma compounds. Correlations were clustered with Ward's two-way hierarchical clustering analysis (HCA). A Pareto-scaled PCA with aroma compound peak area means as independent variables and sensory attribute mean estimates as dependent variables was also conducted using SIMCA version 14.1 (MKS Data Analytics Solutions, Umea, Sweden).

6.3. Results and Discussion

6.3.1. Linear Mixed Model ANOVA: Significant Sensory Attributes

Attributes with variation and mean estimate differences by clone are bitter, buttery, creamy, earthy, fruity, off-flavors other than bitter, potato-like, sweet, and woody flavor notes as well as the related sensory attributes appearance, aroma intensity, mealy texture, and overall quality (Table 6.1). Sensory panelists discerned between cooking methods for creamy and potato-like flavor notes, mealy texture, and overall quality, however, overall quality is the only attribute with significant variation between clone and cooking method interactions for a few clones. Limited significant clone by cooking method interactions suggests that the influence of cooking method on potato flavor profiles is near negligible.

6.3.2. GC-MS Data Processing and ANOVA: Significant Aroma Compounds

Processing produced 539 analyte peaks in 2014 and 583 in 2015. In the 2014 ANOVA with FDR adjustments, 3.90%, 1.67%, and 0.19% of analytes had significant p-values (α=0.05) by clone, cooking method, and their interaction, respectively. Percentages of analytes with significant p-values for analogous ANOVA treatments in 2015 were 77.74%, 30.98%, and 38.40%. Across 20 principal components in PCA, 6.68% of analytes had a least one significant score (α=0.05) in 2014 compared to 10.29% in 2015. The number of tentatively identifiable, non-contaminant compounds in 2014 was 13 and 32 in 2015. A few tentatively identified compounds had ambiguous fragmentation, resulting in subsequent use of generic names (i.e. saturated hydrocarbon 1) in this study.

In 2014 and 2015 ANOVA of aroma analytes, 11 and 26 compounds have significant variation across clones, respectively (Table 6.2). Various hydrocarbon, ether, ester, aldehyde, ketone, halogenic, and nitrogenous compounds are tentatively identified from spectral matching.

The identification of 2 compounds in 2014 and 4 compounds in 2015 are confirmed by retention time matching with standards. Despite some significant analyte variation by cooking method and the interaction of clone by cooking method, scant significance by cooking method and the interaction with clone for sensory attributes in the linear mixed model ANOVA suggests it is not important to overall flavor perception.

6.3.3. Spearman's Rank Correlation Coefficients: Correlations Between Sensory Attributes and Aroma Compounds

Only clone means for sensory attributes and aroma analytes with significant variation across clone are used in combined statistical analysis. Several correlations occur between sensory attributes and aroma compounds (Figure 6.1). The strongest correlation occurs negatively between buttery and 5-methyl-2-hexanone, however, the two do not cluster in subsequent Spearman HCA or in PCA. Other correlations are generally fairly weak, with Spearman's rank correlation coefficients ranging between 0.50 and 0.65 at most.

Since a larger overall quality sensory score implies a positive perception of a potato sample, sensory attributes with a positive Spearman's rank coefficient correlation to overall quality suggest positive flavor attributes (Figure 6.2). Conversely, attributes with a negative correlation to overall quality indicate negative flavor attributes. Positive attributes are appearance, buttery, creamy, fruity, mealy texture, potato-like, and sweet, whereas negative attributes are aroma intensity, bitter, earthy, off-flavors other flavor, and woody.

In HCA clustering of Spearman's rank correlations, two primary clusters emerge (Figure 6.2). Cluster A contains buttery with 1-nonanal (AD-15-3), benzaldehyde (AD-15-1), 2-phenylacetaldehyde (AD-14-5), (E)-2-heptenal (AD-15-2), pentanal (AD-15-4), (Z)-2-methyl-2-penten-1-ol (AC-15-3), 1-pentanol (AC-15-2), 3,4,5-trimethyl-2-cyclopenten-1-one (KE-15-3), 2-ethylfuran (FU-15-1), o-methylacetophenone (KE-14-1), 3,5-octadien-2-one (KE-14-2), (E,E)-2,4-nonadiene (HC-14-1), hexanal (AD-14-2), 2,4-nonadienal (AD-14-3), (E)-2-heptenal (AD-14-1), 3,5-dimethylcyclopentene (HC-14-2), furfural (FU-14-2), and (E)-2-octenal (AD-14-4). The other major cluster, cluster B, contains overall quality, sweet, potato-like, creamy, appearance, mealy texture, fruity, woody, aroma intensity, earthy, off-flavors other than bitter, and bitter with unsaturated hydrocarbon 1 (HC-15-6), 3-methyl-pentane (HC-15-1), saturated hydrocarbon 2 (HC-15-4), 2,2,3,4-tetramethyl-pentane (HC-15-2), 2-formyl-1-methylpyrrole (PR-15), 5-methyl-2-hexanone (KE-15-1), 2-pentyl-furan (FU-15-2), 2-isopropyl-3-methoxypyrazine (PZ-15), 1-octen-3-ol (AC-15-1), p-methylacetophenone (KE-15-2), 3-carene (TE-15-1), α-copaene (TE-14), unidentifiable phenyl acetate (PA-15-2), and α-copaene (TE-15-2). The trend driving separation of cluster A and B is unclear; compound clustering does not reflect characteristics including retention time, atomic mass, density, boiling point, vapor pressure, carbon content, or organoleptic descriptions.

In cluster 1 within cluster A, buttery clusters with 1-nonanal (AD-15-3, r_s=0.56), benzaldehyde (AD-15-1, r_s=0.67), 2-phenylacetaldehyde (AD-14-5, r_s=0.56), (E)-2-heptenal (AD-15-2, r_s=0.28), pentanal (AD-15-4 r_s=0.47), and (Z)-2-methyl-2-penten-1-ol (AC-15-3, r_s=0.60). Because the aroma compounds cluster with buttery flavor, they are likely candidates for buttery flavor biomarkers. Existing organoleptic descriptions of some of the aroma compounds relate with buttery (Table 6.2). Similarly in cluster 2 within cluster B, bitter clusters with 3-carene (TE-15-1, r_s=0.65), α-copaene (TE-14, r_s=0.47, and TE-15-2, r_s=0.63), and an unidentifiable phenyl acetate (PA-15-2, r_s=0.63). Cluster 3 within cluster B contains several sensory attributes and a few aroma compounds: bitter, off-flavors other than bitter, earthy, aroma intensity, and woody with 2-isopropyl-3-methoxypyrazine (PZ-15), 1-octen-3-ol (AC-15-1), and p-methylacetophenone (KE-15-2). Each aroma compound in cluster 3 is characterized by earthy (including vanilla) organoleptic descriptions. Mealy texture also positively clusters with saturated hydrocarbon 2 (HC-15-4, r_s=0.53) and 2,2,3,4-tetramethyl-pentane (HC-15-2, r_s=0.50) in cluster 4 of cluster B.

6.3.4. Principle Component Analysis: Multivariate Evaluation of the Relationship Between Sensory Attributes and Aroma Compounds

The PCA of analyte and sensory attribute means by clone has two principal components (PC) that capture 49.5% (R^2) of clonal variation, although Q^2 validation is only 17.8%. A cumulative Q^2 value disproportionally smaller than R^2 suggests a poor model fit, yet values for some individual analyte and sensory attribute loadings for either principal component are more proportional (Figure 6.3). In principal component 1, bitter, off-flavors other than bitter, earthy, and benzaldehyde (AD-15-1) are modeled well as demonstrated by large R^2 and Q^2 values (>0.50). More loadings are modeled well by principal component 2, including (E)-2-heptenal (AD-14-1), 3,5-dimethylcyclopentene (HC-14-2), (E)-2-octenal (AD-14-4), 2-n-butyl furan (FU-14-1), (E,E)-2,4-nonadiene (HC-14-1), 2,4-nonadienal (AD-14-3), hexanal (AD-14-2), o-methylacetophenone (KE-14-2), and 3,5-octadien-2-one (KE-14-2), all of which were identified from 2014 data.

Two primary clusters occur in a loading and score normalized correlation biplot of the PCA (Figure 6.4). In cluster 1, sweet, potato-like, buttery, overall quality, and creamy, all positive attributes according to Spearman's rank coefficient correlation, correlate positively together with pentanal (AD-15-4), benzaldehyde (AD-15-1), (E)-2-heptenal (AD-15-2), benzoate-3-methyl-2-buten-1-ol (PA-15-1), 2-ethylfuran (FU-15-1), 3,4,5-trimethyl-2-cyclopenten-1-one (KE-15-3), (Z)-2-methyl-2-penten-1-ol (AC-15-3), and 1-nonanal (AD-15-3).

Half-sib yellow-fleshed cultivars Masquerade and Harvest Moon as well as white-fleshed cultivar Russet Nugget correlate to cluster 1.

The opposite cluster towards the right of the PCA biplot, cluster 2, contains the negative attributes earthy, woody, bitter, aroma intensity, and off-flavors other than bitter correlated with 3-methyl-pentane (HC-15-1) and 2-isopropyl-3-methoxypyrazine (PZ-15-1). In HCA, the same sensory attributes cluster with 2-isopropyl-3-methoxypyrazine. Red-fleshed clones CO04063-4R/R and Crimson King correlate with the cluster, as well as purple-fleshed clone CO04056-3P/PW to a lesser extent. White-fleshed clones AC00395-2RU, Russet Norkotah, Russet Burbank, and Fortress Russet plus yellow-fleshed clone CO04067-8R/Y and purple-fleshed clone Purple Majesty plot towards the middle of the biplot, indicating that the clones are poorly described by sensory attributes or analytes. Mealy texture and a few other analytes are also located towards the middle of the plot, implying minimal differences between clones for those variables. Appearance is between the two primary clusters, indicating that it is not correlated to either cluster, though it does weakly correlate to yellow-fleshed clone CO04099-3W/Y. Clone CO04099-3W/Y has an appearance different than that of the other clones, though it is unknown if it is better or worse in comparison.

6.3.5. Application of Results

Based on Spearman's rank coefficient correlations with overall quality, positive versus negative attributes are revealed. Positive attributes are appearance, buttery, creamy, fruity, mealy texture, potato-like, and sweet, which should be maximized to increase flavor quality. Negative attributes, which should be minimized, are aroma intensity, bitter, earthy, off-flavors other flavor, and woody.

Results provide an overview of cooked flavor for clones, particularly those developed by the CSU Breeding and Selection program in relation to common commercial cultivars. About half of the potato clones show variation in PCA amongst sensory attribute and aroma compound clusters, whereas remaining clones are not very different. Masquerade and Harvest Moon, half-sib, yellow-fleshed clones developed by the CSU Breeding and Selection program, correlate with a cluster of positive sensory attributes, indicating favorable flavor reception compared to the common yellow-fleshed cultivar Yukon Gold, which is not distinguished by positive or negative attributes. Yellow-fleshed clones CO04067-8R/Y, CO04099-3W/Y, and Red Luna are comparable to Yukon Gold. Russet Nugget was distinguished by positive attributes compared to the common, undistinguished russet cultivar, Russet Burbank. Other russet clones AC00395-2RU, Fortress Russet, and Russet Norkotah are comparable to Russet Burbank. Purple-fleshed advanced selection CO04056-3P/PW as well as red-fleshed clones CO04063-4R/R and Crimson King correlate to negative flavor attributes. Purple Majesty, which is a commercially successful purple-fleshed cultivar, is not distinguished by positive or negative attributes, suggesting the formerly mentioned purple- and red-fleshed may have poor flavor quality.

In terms of potential biomarkers, aroma compounds that consistently coincided with the positive buttery sensory attribute in Spearman's rank correlation HCA and PCA are aldehydes 1-nonanal, benzaldehyde, (E)-2-heptenal, pentanal, 2-phenylacetaldehyde, and the alcohol (Z)-2-methyl-2-penten-1-ol. Other potential biomarkers for positive flavor attributes, identified through either HCA or PCA, are benzoate-3-methyl-2-buten-1-ol, 2-ethylfuran, and 3,4,5-trimethyl-2-cyclopenten-1-one. The aroma compound 2-isopropyl-3-methoxypyrazine consistently correlates with negative sensory attributes, making it a negative flavor biomarker candidate. Based on HCA, terpenes are also potential negative flavor biomarkers, particularly for bitter.

To confirm feasibility of biomarkers, additional quantitative analysis should be carried out to assess clonal variation. Aroma compounds without variation between clones are poor biomarkers for selective breeding, which depends on heterogeneity for trait improvement. Correlations of clones in this study suggest some degree of heterogeneity exists for proposed biomarkers. Clone correlations or lack of correlations in the case of white-fleshed russets imply that flesh color is somewhat indicative of cooked flavor. Color-fleshed potatoes tend to contain larger amounts of secondary metabolite anthocyanins, carotenoids, and other phytopigments compared to white-fleshed potatoes, supporting this distinction. Perhaps secondary metabolites serve as reactants for some aroma compound formation reactions.

6.4. Summary

In this study, aroma compound biomarkers for cooked potato flavor were determined. Sensory analysis was paired with a non-targeted gas chromatography-mass spectrometry (GC-MS) aroma assay to evaluate fifteen potato clones. Spearman's rank coefficient correlations suggested that positive sensory attributes were appearance, buttery, creamy, fruity, mealy texture, potato-like, and sweet, whereas negative attributes were aroma intensity, bitter, earthy, off-flavors other flavor, and woody. Aroma compounds that consistently coincided with buttery in Spearman's rank correlation HCA and PCA were aldehydes 1-nonanal, benzaldehyde, (E)-2-heptenal, pentanal, 2-phenylacetaldehyde, and the alcohol (Z)-2-methyl-2-penten-1-ol, all of which are potential biomarkers for buttery. Other potential biomarkers for positive flavor attributes are benzoate-3-methyl-2-buten-1-ol, 2-ethylfuran, and 3,4,5-trimethyl-2-cyclopenten-1-one. The aroma compound 2-isopropyl-3-methoxypyrazine consistently correlated with negative sensory attributes, making it a negative flavor biomarker candidate. Terpenes are also potential negative flavor biomarkers, particularly for bitter.

Table 6.1. Cooked potato linear mixed model ANOVA[a] of sensory attributes rated by a trained sensory panel

Attribute Type	Attribute	Clone ANOVA F-test		Cooking Method ANOVA F-test		Clone:Cooking Method ANOVA F-test	
Flavor Note	Bitter	$F_{14,179}$ =5.484, p =0.000	*	$F_{1,347}$ =0.435, p =0.510		$F_{14,356}$ =1.430, p =0.136	
	Buttery	$F_{14,497}$ =3.845, p =0.000	*	$F_{1,500}$ =2.421, p =0.120		$F_{14,482}$ =1.616, p =0.071	
	Creamy	$F_{14,486}$ =2.626, p =0.001	*	$F_{1,11}$ =11.286, p =0.007	*	$F_{14,473}$ =1.674, p =0.058	
	Earthy	$F_{14,484}$ =4.387, p =0.000	*	$F_{1,9}$ =1.025, p =0.339		$F_{14,470}$ =0.392, p =0.977	
	Fruity	$F_{14,498}$ =2.208, p =0.007	*	$F_{1,502}$ =1.594, p =0.207		$F_{14,483}$ =1.049, p =0.403	
	Lemon	$F_{14,484}$ =1.657, p =0.061		$F_{1,10}$ =0.690, p =0.426		$F_{14,471}$ =1.032, p =0.420	
	Off-Flavors Other Than Bitter	$F_{14,499}$ =3.778, p =0.000	*	$F_{1,511}$ =0.583, p =0.445		$F_{14,483}$ =1.271, p =0.221	
	Potato-Like	$F_{14,490}$ =2.750, p =0.001	*	$F_{1,13}$ =16.479, p =0.001	*	$F_{14,476}$ =0.722, p =0.753	
	Sweet	$F_{14,484}$ =3.556, p =0.000	*	$F_{1,9}$ =0.084, p =0.779		$F_{14,470}$ =0.552, p =0.901	
	Umami	$F_{14,190}$ =0.622, p =0.845		$F_{1,12}$ =1.914, p =0.193		$F_{14,362}$ =0.896, p =0.564	
	Woody	$F_{14,484}$ =2.784, p =0.001	*	$F_{1,10}$ =0.597, p =0.458		$F_{14,471}$ =0.763, p =0.709	
Other	Appearance	$F_{14,170}$ =4.022, p =0.000	*	$F_{1,3}$ =0.376, p =0.583		$F_{14,324}$ =0.786, p =0.685	
	Aroma Intensity	$F_{14,481}$ =2.235, p =0.006	*	$F_{1,8}$ =0.106, p =0.753		$F_{14,468}$ =0.688, p =0.787	
	Mealy Texture	$F_{14,487}$ =5.004, p =0.000	*	$F_{1,12}$ =14.18, p =0.003	*	$F_{14,473}$ =1.015, p =0.436	
	Overall Quality	$F_{14,149}$ =6.719, p =0.000	*	$F_{1,14}$ =4.955, p =0.044	*	$F_{14,334}$ =2.444, p =0.003	*

[a]Means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=8-21 hedonic scores x 15 clones x 2 cooking methods). Sensory attributes with asterisks have significant variation according to mixed model ANOVA F-tests (α=0.05), confirmed by at least one significant difference between means according to linear least square mean contrasts with Welch's adjustments (t-test, α=0.05).

Table 6.2. Non-targeted HS-SPME GC/MS detection of cooked potato analytes with significant variation by clone[a]

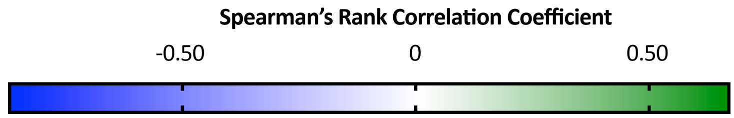
Year	Primary Functional Group	Configuration	Compound Code[b]	Compound[c]	Organoleptics[d]	Top 5 Mass Peaks	RT (s)	Clone ANOVA F-test
2014	**Hydrocarbon**	**Alkene**	HC-14-1	(E,E)-2,4-nonadiene		79, 81, 53, 95, 68	624	F_{14} =4.531, p =0.000
		Cycloalkene	HC-14-2	3,5-dimethylcyclopentene		81, 79, 41, 53, 39	749	F_{14} =6.166, p =0.000
		Terpene	TE-14	α-copaene*	woody, spicy, honey	105, 119, 91, 161, 93	1452	F_{14} =5.472, p =0.000
	Aldehyde	**Aliphatic**	AD-14-1	(E)-2-heptenal	green, fatty, fruity	41, 83, 55, 39, 69	649	F_{14} =5.857, p =0.000
			AD-14-2	hexanal	green, woody, fatty	41, 44, 56, 43, 57	348	F_{14} =6.467, p =0.000
			AD-14-3	2,4-nonadienal	fatty, nutty, citrus	81, 41, 67, 39, 65	1172	F_{14} =7.182, p =0.000
			AD-14-4	(E)-2-octenal	green, herbal, fatty	41, 55, 83, 70, 39	839	F_{14} =5.802, p =0.000
			AD-14-5	2-phenylacetaldehyde*	honey, floral, cocoa	91, 92, 65, 93, 63	810	F_{14} =2.834, p =0.001
	Ketone	**Aliphatic**	KE-14-1	o-methylacetophenone	floral, burnt, nutty	91, 79, 105, 134, 119	1063	F_{14} =3.815, p =0.000
			KE-14-2	3,5-octadien-2-one	fruity, green	95, 79, 81, 71, 59	914	F_{14} =4.119, p =0.000
	Ether	**Furan**	FU-14-1	2-n-butyl furan	fruity, wine, spicy	81, 67, 95, 70, 82	1363	F_{14} =5.311, p =0.000
2015	**Hydrocarbon**	**Alkane**	HC-15-1	3-methyl-pentane		57, 56, 53, 50, 51	226	F_{13} =16.91, p =0.000
			HC-15-2	2,2,3,4-tetramethyl-pentane		57, 41, 43, 56, 71	818	F_{13} =5.047, p =0.000
			HC-15-3	saturated hydrocarbon 1		57, 41, 43, 71, 70	758	F_{13} =4.356, p =0.000
			HC-15-4	saturated hydrocarbon 2		57, 41, 56, 71, 70	795	F_{13} =4.986, p =0.000
			HC-15-5	saturated hydrocarbon 3		57, 41, 56, 71, 68	934	F_{13} =6.042, p =0.000
		Alkene	HC-15-6	unsaturated hydrocarbon 1		57, 56, 55, 53, 61	144	F_{13} =4.892, p =0.000
			HC-15-7	unsaturated hydrocarbon 2		57, 41, 56, 55, 69	845	F_{13} =3.830 p =0.000
		Terpene	TE-15-1	3-carene*	citrus, pine	93, 67, 121, 81, 53	732	F_{13} =21.520, p =0.000
			TE-15-2	α-copaene*	woody, spicy, honey	161, 119, 149, 204, 145	1437	F_{13} =14.210, p =0.000
	Alcohol	**Aliphatic**	AC-15-1	1-octen-3-ol*	earthy, green, fatty	57, 69, 55, 67, 81	674	F_{13} =18.000, p =0.000
			AC-15-2	1-pentanol	fermented, yeasty, wine	41, 55, 42, 71, 70	276	F_{13} =4.955, p =0.000
			AC-15-3	(Z)-2-methyl-2-penten-1-ol		72, 71, 67, 43 57	332	F_{13} =16.930, p =0.000
	Aldehyde	**Aliphatic**	AD-15-1	benzaldehyde	nutty, fruity, fatty	77, 106, 51, 50, 52	642	F_{13} =9.435, p =0.000
			AD-15-2	(E)-2-heptenal	green, fatty, fruity	41, 55, 57, 39, 69	632	F_{13} =5.628, p =0.000
			AD-15-3	1-nonanal	citrus, green, potato	41, 57, 43, 55, 56	912	F_{13} =3.982, p =0.000
			AD-15-4	pentanal	fermented, yeasty, wine	44, 58, 43, 41, 57	172	F_{13} =7.753, p =0.000
	Ketone	**Aliphatic**	KE-15-1	5-methyl-2-hexanone		43, 58, 81, 71, 82	503	F_{13} =3.330, p =0.000
		Acetophenone	KE-15-2	p-methylacetophenone	creamy, fruity, vanilla	91, 119, 92, 134, 65	936	F_{13} =6.124, p =0.000
		Cyclic	KE-15-3	3,4,5-trimethyl-2-cyclopenten-1-one		109, 81, 79, 124, 53	428	F_{13} =7.328, p =0.000
	Ether	**Furan**	FU-15-1	2-ethylfuran	musty, earthy, yeasty	81, 53, 96, 65, 82	171	F_{13} =4.526, p =0.000
			FU-15-2	2-pentyl-furan	fruity, green, earthy	82, 81, 53, 138, 39	686	F_{13} =4.901, p =0.000
	Ester	**Phenyl Acetate**	PA-15-1	benzoate-3-methyl-2-buten-1-ol	woody, fruity, chocolate	105, 41, 43, 55, 69	716	F_{13} =3.967, p =0.000
		Phenyl Acetate	PA-15-2	unidentifiable phenyl acetate		105, 91, 79, 93, 106	1435	F_{13} =7.430, p =0.000
	Halogenic	**Aliphatic**	HA-15	1-chloro-2-methyl-butane		57, 41, 55, 39, 56	158	F_{13} =1.536, p =0.000
	Nitrogenous	**Pyrazine**	PZ-15	2-isopropyl-3-methoxypyrazine*	earthy, chocolate, nutty	109, 152, 137, 124, 105	883	F_{13} =5.268, p =0.000
		Pyrrole	PR-15	2-formyl-1-methylpyrrole		108, 109, 42, 40, 95	546	F_{13} =4.254, p =0.000

[a]Analytes have significant variation by clone according to ANOVA F-tests (α=0.05), confirmed by at least one significant difference between means according to Tukey's HSD t-tests (α=0.05) (n=10 x 14 clones).

[b]Compound codes are arbitrary, but were intended simplify table or figure labels as appropriate; The first letters are AC=alcohol, AD=aldehyde, FU=furan, HA= halogenic, HC=hydrocarbon, KE=ketone, PA=phenyl acetate, PR=pyrrole, PZ=pyrazine, SU=sulfurous, and TE=terpene; The second number digits represent growing season of potato material, 14=2014, 15=2015; Final number digit makes each code unique per analyte.

[c]Compounds were tentatively identified using spectral matching; identifications of compounds with an asterisks were confirmed with gas chromatography retention time matching.

[d]Organoleptic characteristics are from the Good Scents Company online database.



Class	Subclass	Analyte	Appearance	Aroma Intensity	Bitter	Buttery	Creamy	Earthy	Fruity	Mealy Texture	Other Off-Flavors	Overall Quality	Potato-Like	Sweet	Woody
Hydrocarbon		(E,E)-2,4-nonadiene	-0.07	-0.27	0.13	0.08	-0.21	-0.38	0.20	-0.12	-0.03	-0.24	-0.11	-0.13	-0.28
Hydrocarbon		3,5-dimethylcyclopentene	-0.15	-0.45	-0.07	0.15	-0.31	-0.44	0.06	-0.16	-0.10	-0.26	0.02	-0.03	-0.33
Hydrocarbon		3-methyl-pentane	0.09	0.14	0.15	-0.08	0.01	0.51	0.25	-0.09	0.16	-0.09	-0.13	0.27	0.29
Hydrocarbon		2,2,3,4-tetramethyl-pentane	0.47	0.12	0.27	-0.06	0.04	0.35	0.08	0.50	0.05	0.26	0.00	-0.06	0.37
Hydrocarbon		saturated hydrocarbon 2	0.49	0.09	0.24	-0.07	0.00	0.32	0.11	0.53	0.01	0.29	0.01	-0.01	0.34
Hydrocarbon		unsaturated hydrocarbon 1	0.15	0.04	-0.20	0.14	0.21	0.33	-0.11	0.36	-0.16	0.36	0.02	0.21	0.29
Hydrocarbon	Terpene	3-carene	0.02	0.52	0.65	-0.45	0.16	0.63	0.12	-0.14	0.39	-0.32	-0.18	-0.58	0.31
Hydrocarbon	Terpene	α-copaene (2014)	0.15	0.32	0.47	-0.12	0.03	0.22	0.30	-0.15	0.15	-0.07	-0.02	-0.14	0.19
Hydrocarbon	Terpene	α-copaene (2015)	0.12	0.37	0.63	-0.32	0.01	0.34	0.11	-0.06	0.28	-0.24	0.06	-0.43	0.20
Alcohol		1-octen-3-ol	-0.22	0.47	0.65	-0.44	-0.28	0.35	0.44	-0.25	0.61	-0.40	-0.49	-0.03	0.47
Alcohol		1-pentanol	-0.26	-0.09	-0.01	-0.31	-0.25	-0.08	-0.14	-0.49	0.09	-0.36	-0.20	-0.21	-0.11
Aldehyde		(E)-2-heptenal (2014)	-0.15	-0.38	-0.12	0.12	-0.39	-0.35	0.12	-0.14	-0.13	-0.15	0.11	0.11	-0.23
Aldehyde		(E)-2-heptenal (2015)	0.09	-0.32	-0.14	0.28	0.29	-0.21	0.07	-0.33	-0.27	-0.05	-0.17	0.02	-0.58
Aldehyde		hexanal	-0.12	-0.55	-0.11	0.15	-0.37	-0.43	-0.05	-0.07	-0.16	-0.24	0.04	-0.04	-0.30
Aldehyde		2,4-nonadienal	-0.06	-0.51	-0.16	0.26	-0.22	-0.44	0.04	-0.14	-0.19	-0.19	0.09	0.05	-0.42
Aldehyde		(E)-2-octenal	-0.15	-0.51	-0.12	0.11	-0.39	-0.43	0.06	-0.15	-0.17	-0.25	0.03	0.03	-0.36
Aldehyde		2-phenylacetaldehyde	0.15	-0.38	-0.24	0.56	0.52	-0.45	0.26	-0.24	-0.31	0.05	-0.18	0.21	-0.60
Aldehyde		(Z)-2-methyl-2-penten-1-ol	0.50	-0.40	-0.31	0.60	0.46	-0.28	-0.01	0.15	-0.52	0.35	0.16	0.10	-0.59
Aldehyde		benzaldehyde	0.18	-0.28	-0.32	0.67	0.56	-0.44	0.13	-0.04	-0.39	0.27	-0.17	0.20	-0.65
Aldehyde		1-nonanal	0.21	-0.09	-0.18	0.56	0.61	-0.30	0.07	-0.09	-0.34	0.24	-0.17	0.07	-0.48
Aldehyde		pentanal	0.15	-0.12	-0.28	0.47	0.47	-0.20	-0.32	0.01	-0.25	0.25	0.10	-0.20	-0.27
Ketone		o-methylacetophenone	0.00	-0.27	0.03	0.04	-0.20	-0.42	0.27	0.08	-0.13	-0.17	0.00	0.02	-0.40
Ketone		3,5-octadien-2-one	-0.09	-0.49	-0.07	0.13	-0.23	-0.30	0.08	-0.03	-0.06	-0.27	-0.12	-0.02	-0.27
Ketone		5-methyl-2-hexanone	-0.27	0.27	0.43	-0.87	-0.68	0.34	0.22	0.10	0.35	-0.41	-0.17	-0.33	0.28
Ketone		p-methylacetophenone	-0.17	0.34	0.55	-0.28	-0.14	0.48	0.21	-0.62	0.58	-0.65	-0.27	-0.06	0.43
Ketone		3,4,5-trimethyl-2-cyclopenten-1-one	-0.22	-0.27	-0.08	-0.16	-0.26	-0.45	0.02	-0.37	-0.16	-0.26	0.05	-0.04	-0.53
Furan		furfural	-0.18	-0.48	-0.05	0.03	-0.43	-0.40	0.02	-0.19	-0.11	-0.33	0.04	-0.02	-0.36
Furan		2-ethylfuran	-0.19	-0.03	0.09	-0.16	-0.22	-0.04	-0.21	-0.26	0.17	-0.38	0.12	-0.44	-0.12
Furan		2-pentyl-furan	0.08	0.35	0.31	-0.52	-0.26	0.35	0.30	-0.04	0.13	-0.18	0.06	-0.18	0.10
		unidentifiable phenyl acetate	0.13	0.36	0.63	-0.31	0.03	0.33	0.11	-0.06	0.27	-0.23	0.02	-0.44	0.19
Nitrogenous		2-isopropyl-3-methoxypyrazine	0.15	0.65	0.46	-0.59	-0.33	0.53	0.42	0.08	0.34	0.01	-0.09	-0.02	0.51
Nitrogenous		2-formyl-1-methylpyrrole	-0.09	0.57	0.18	-0.21	-0.03	0.36	0.22	-0.16	0.19	-0.10	0.10	-0.11	0.13

Figure 6.1. Spearman's rank correlation coefficients for sensory attributes[a] and aroma analytes[b] of cooked potatoes

[a]Clone means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=17-38 hedonic scores x 15 clones).

[b]Log-transformed peak area means detected by HS-SPME GC-MS (n=10 technical replicates x 15 clones per year).

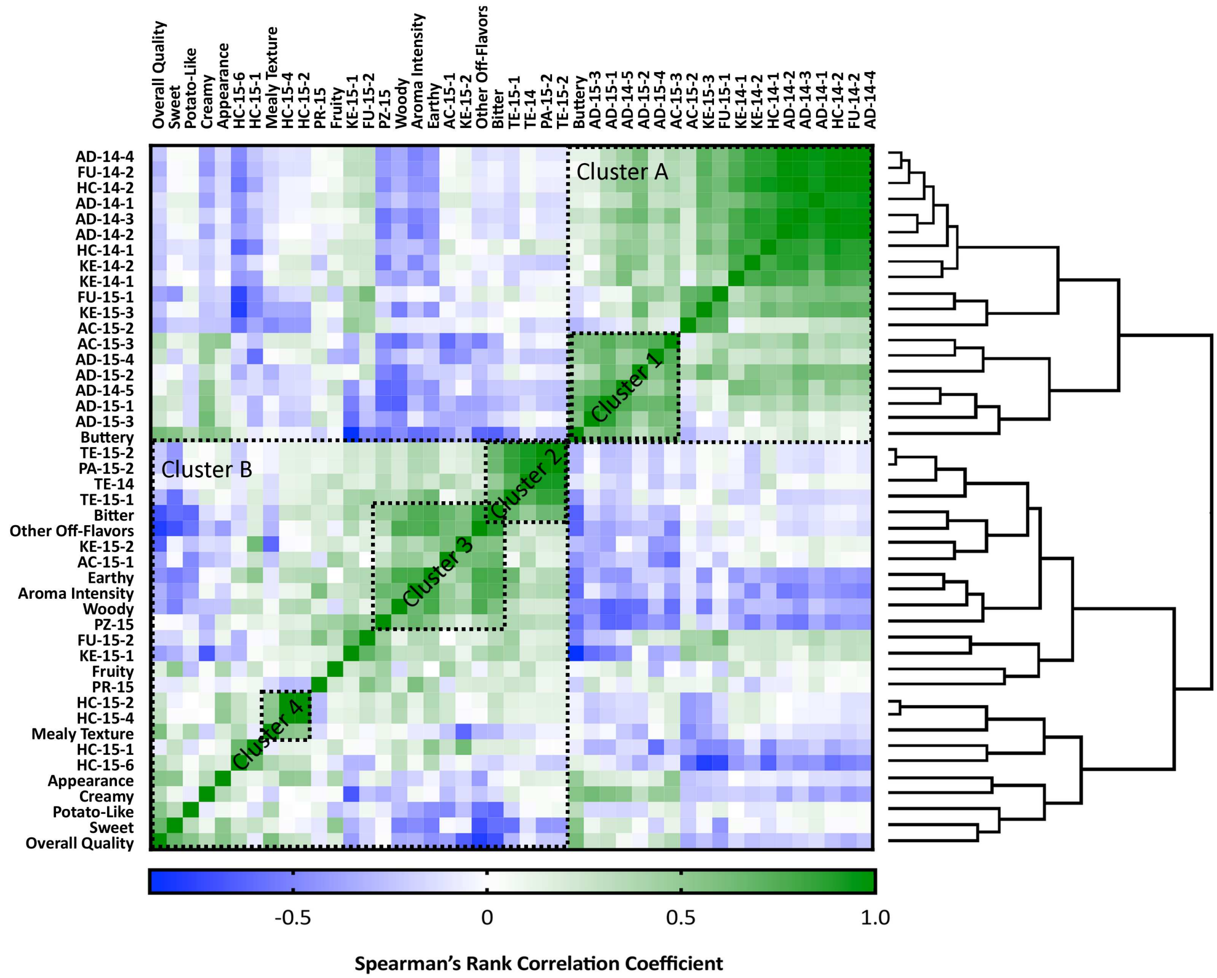


Figure 6.2. Spearman's rank correlation coefficients with two-way Ward HCA for sensory attributes[a] and aroma analytes[b] of cooked potatoes

[a]Clone means estimated using REML in a mixed model analysis with session year, sample order presented to panelists, panelists, interactions as random effects and clone, cooking method, and the interaction between clone and cooking method as fixed effects (n=17-38 hedonic scores x 15 clones).

[b]Log-transformed peak area means detected by HS-SPME GC-MS (n=10 technical replicates x 15 clones per year).

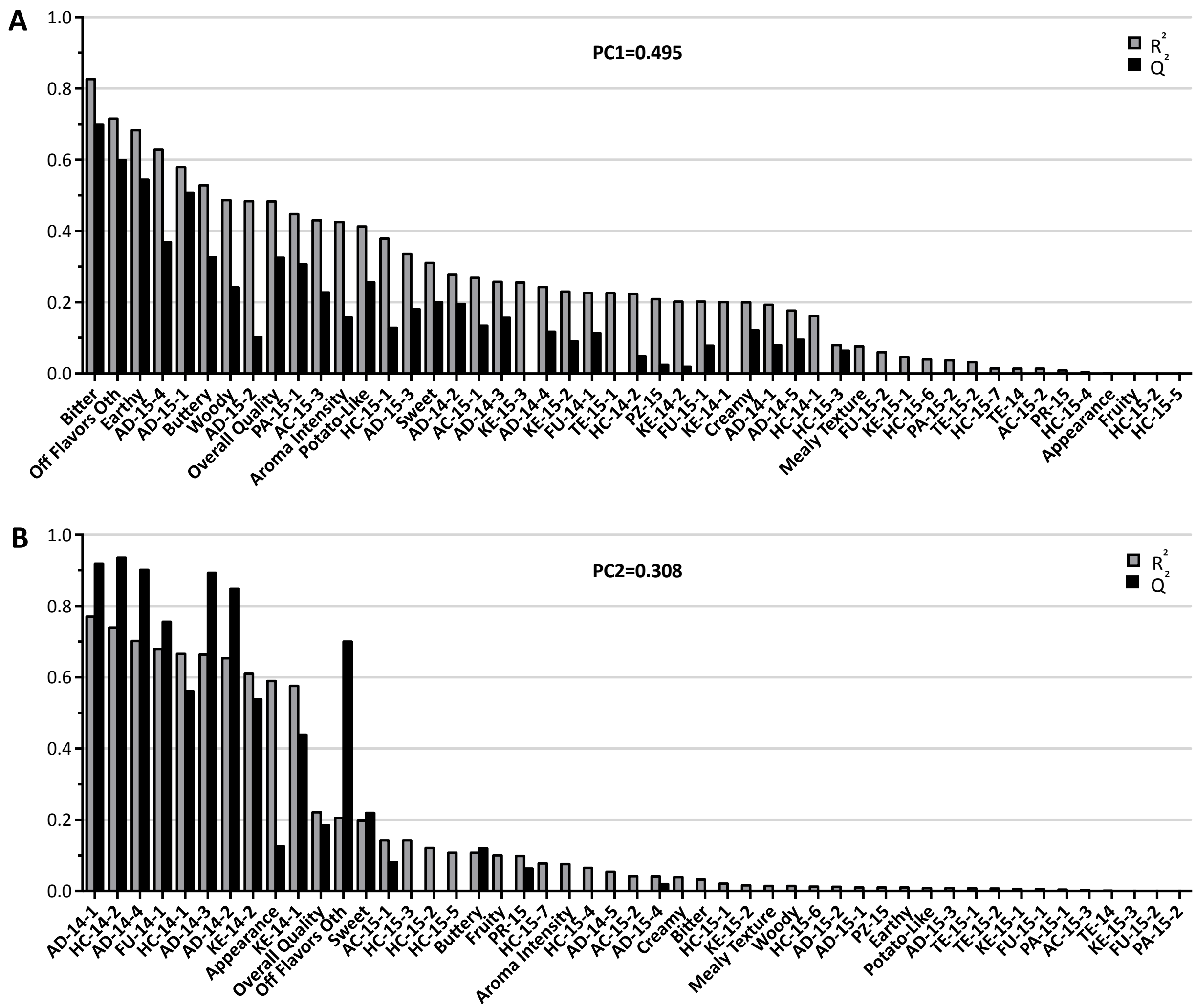


Figure 6.3. Contribution plots for principle component analysis of Pareto-scaled sensory attribute[a] and analyte[b] loadings

[a]All sensory attributes are significantly different according to mixed model ANOVA F-tests (α=0.05) for clone (n=8-21 hedonic scores x 2 cooking methods x 15 clones).

[b]All analytes are significantly different according to ANOVA F-tests (α=0.05) for clone (n=10 technical replicates x 15 clones per year).

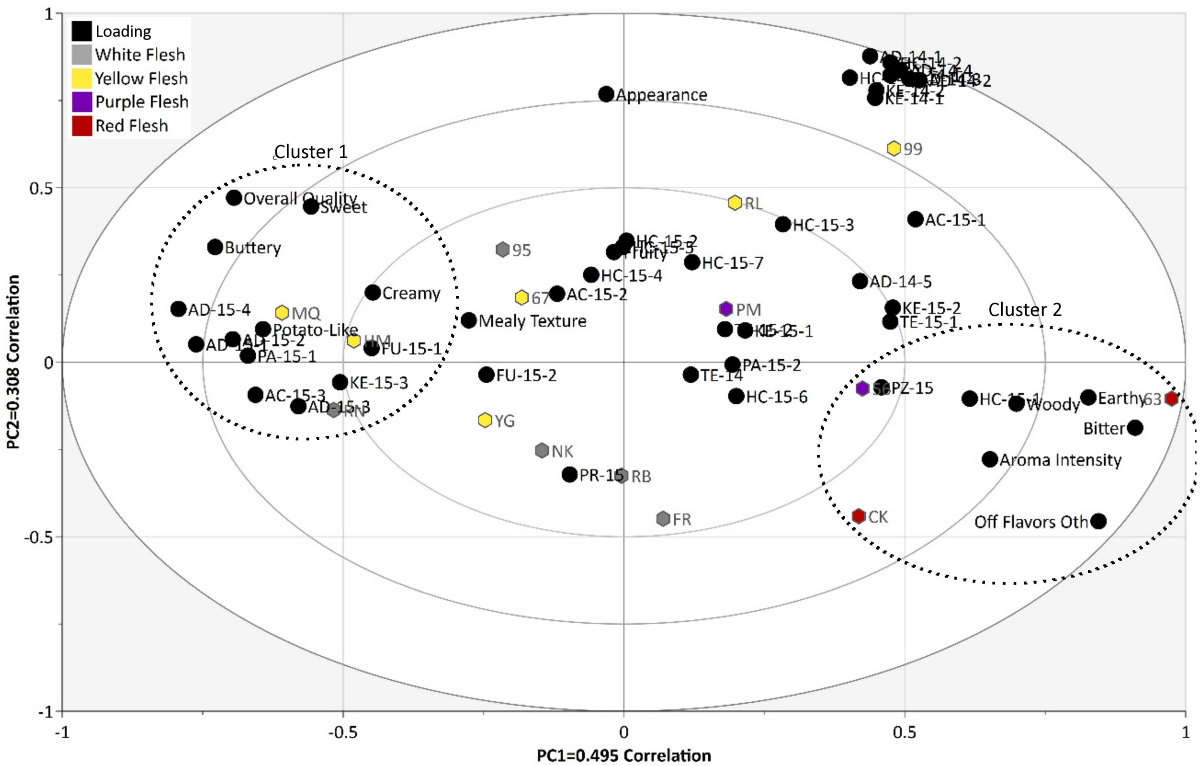


Figure 6.4. Normalized correlation R^2 biplot in principle component analysis of Pareto-scaled sensory attribute[a] and analyte[b] loadings and potato clone[c] scores colored by flesh color

[a]All sensory attributes are significantly different according to mixed model ANOVA F-tests (α=0.05) for clone (n=8-21 hedonic scores x 2 cooking methods x 15 clones).

[b]All analytes are significantly different according to ANOVA F-tests (α=0.05) for clone (n=10 technical replicates x 15 clones per year).

[c]95=AC00395-2RU, FR=Fortress Russet, RB=Russet Burbank, NK=Russet Norkotah, RN=Russet Nugget, 67=CO04067-8R/Y, 99=CO04099-3W/Y, HM=Harvest Moon, MQ=Masquerade, RL=Red Luna, YG=Yukon Gold, 56=CO04056-3P/PW, PM=Purple Majesty, 63=CO04063-4R/R, CK=Crimson King.

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CHAPTER 7. CONCLUSION

Sensory data suggests two opposing groups of attributes: positive attributes, where high hedonic scores are desirable, and negative attributes, which should have lower hedonic scores. Overall quality scores identify positive versus negative attributes, depending on respective proportional or inverse relationships. Positive attribute scores are proportional to overall quality scores, whereas negative attribute scores are inversely related to overall quality. Consistent positive attributes are appearance, buttery, creamy, fruity, sweet, overall quality, and potato-like flavor, whereas consistent negative attributes are aroma intensity, bitter, earthy, off-flavors other than bitter, and woody. Fewer sensory attributes in combined sensory and volatile metabolite analyses appear to be important to potato flavor. Positive attributes in combined analysis are buttery, sweet, and potato-like flavor and negative attributes are earthy, woody, aroma intensity, bitter, and off-flavors other than bitter. Sensory attribute profiles differ by clone.

Cooking method plays a role for some potato volatile metabolites and sensory attributes, however, interactions between clones and cooking methods are nearly absent in sensory analysis, implying a limited genetic basis. Clone differences for sensory attributes, volatile metabolites, and clonal correlations in combined analysis suggests some degree of heritability of positive or negative flavor traits, as well as existing heterogeneity for those traits. Sensory perceptions seem to be related to flesh color in addition to clone. Sensory scores are more consistent for white-fleshed potatoes compared to color-fleshed, most likely due to greater breeding emphasis on white-fleshed russet potatoes, although the appearance of colored flesh is consistently favored over white flesh. Because red-fleshed potatoes in particular have received less attention in breeding, less selection against bitterness or other off-flavors caused by phytopigments, glycoalkaloids, or other secondary plant metabolites may be related to flavor inconsistencies and off-flavor compound abundance.

Similarly to other studies, a large number of identified potato aroma compounds are aldehydes. The prominence of aldehydes and furans, which contain aldehyde functional groups, in significant clone by cooking method interactions suggests formation dependence on cooking, most likely through Maillard or Strecker reactions. Other identified aroma compounds have greater abundance in baked versus boiled potatoes. Since baked potatoes are subject to higher cooking temperatures and drier conditions, greater abundance of aroma compounds supports consensus of the importance of thermal reactions during cooking for aroma compound formation. Aroma compound profiles are different by clone.

Sensory analysis attribute scores for mealy texture and texture instrumental analysis of hardness are not correlated. Sensory mealy textures scores are variable by clone but instrumental measurements are not. Other instrumental texture measurements may be more appropriate to measure mealy texture. Sensory texture differences by clone imply a genetic component for texture. Differences in hardness for cooking method reveal an environmental component for cooked potato texture as well.

Several aroma compounds are identified as potential biomarkers of cooked potato flavor in combined sensory and aroma compound analyses for positive sensory attributes: buttery, sweet, and potato-like flavor and negative sensory attributes earthy, woody, aroma intensity, bitter, and off-flavors other than bitter. Potential buttery biomarkers are 1-nonanal, benzaldehyde, (E)-2-heptenal, pentanal, 2-phenylacetaldehyde, and (Z)-2-methyl-2-penten-1-ol, and 5-methyl-2-hexanone. Other potential biomarkers for positive flavor attributes, including buttery, are benzoate-3-methyl-2-buten-1-ol, 2-ethylfuran, and 3,4,5-trimethyl-2-cyclopenten-1-

one. Negative flavor attributes are earthy, woody, aroma intensity, bitter, and off-flavors other than bitter, with 2-isopropyl-3-methoxypyrazine as a potential biomarker. Terpenes (i.e. 3-carene, α-copaene) may also be biomarkers for bitter.

In terms of flavor by clone, Masquerade and Harvest Moon have consistently positive attributes in sensory and combined sensory and aroma compound analyses. The CSU Potato Breeding & Selection Program developed both cultivars, which are actually half-sibs, therefore similarities between the two are not surprising. Fortress Russet and Russet Nugget are comparable to Russet Norkotah and Russet Burbank for flavor. Advanced selection AC00395-2RU is comparable to the other white-fleshed russets as well. Masquerade and Harvest Moon are more favorable for flavor than the common yellow-fleshed cultivar Yukon Gold. Red Luna, CO04099-3W/Y, and CO04067-8R/Y are comparable to Yukon Gold. Advanced selections CO04056-3P/PW, CO04063-4R/R, and Crimson King are associated with a negative flavor compared to Purple Majesty.

Plant breeding efforts tend to focus on yield, postharvest storage, and pest resistance, which is thought to have a negative influence on produce quality, including flavor. Furthermore, flavor is a complex trait that is difficult to select for. In this study, critical aroma compounds are identified by combined sensory and aroma compound analyses. Aroma compounds are candidates for flavor biomarkers, which may facilitate aroma phenotypic selection for improving potato flavor after biomarker confirmation in quantitative assays and development of more efficient, high-throughput methods. Identified aroma compounds may also facilitate discovery of flavor metabolic pathways or genes. Flavor profiles generated from sensory analysis are further informative to potato breeders, producers, and consumers that wish to maximize flavor quality.

APPENDIX 1. SENSORY ANALYSIS SCORECARD

Sensory Panel Session #X, xxth Month, 20xx

Name ________________________________ Order______ Sample Code ______________

Instructions: Print your name, sample order number, and the sample code. **Taste samples by ascending order numbers (1, 2, 3, etc.).** Before tasting a potato sample, first rate the sample for appearance and aroma intensity. Then, you may taste the sample and rate each flavor component as well as mealy texture and overall quality. Place an "x" or checkmark in an empty box under a scale to provide a single rating for each individual characteristic. **Remember to inhale through your mouth or nose and then exhale through your nose while chewing to maximize your flavor perception. Please be sure to rinse your palate between potato samples with the provided crackers and water.** Reference foods for flavors have been provided and may be used as a reminder for individual flavors, but do not necessarily correspond to specific points on rating scales.

Appearance

1 Not appealing	2 Trace of appeal	3 Faintly appealing	4 Slightly appealing	5 Moderately appealing	6 Definitely appealing	7 Strongly appealing	8 Very strongly appealing	9 Extremely appealing

Aroma Intensity

1 No aroma	2 Trace aromatic	3 Faintly aromatic	4 Slightly aromatic	5 Moderately aromatic	6 Definitely aromatic	7 Strongly aromatic	8 Very strongly aromatic	9 Extremely aromatic

Potato-like Flavor

1 Not potato-like	2 Trace potato-like	3 Faintly potato-like	4 Slightly potato-like	5 Moderately potato-like	6 Definitely potato-like	7 Strongly potato-like	8 Very strongly potato-like	9 Extremely potato-like

Sweetness

1 Not Sweet	2 Trace sweet	3 Faintly sweet	4 Slightly sweet	5 Moderately sweet	6 Definitely sweet	7 Strongly sweet	8 Very strongly sweet	9 Extremely sweet

Remember to inhale through your mouth or nose and then exhale through your nose while chewing to maximize your flavor perception. Please be sure to rinse your palate between potato samples with the provided crackers and water.

Fruity Flavor

1 Not fruity	2 Trace fruity	3 Faintly fruity	4 Slightly fruity	5 Moderately fruity	6 Definitely fruity	7 Strongly fruity	8 Very strongly fruity	9 Extremely fruity

Lemon Flavor

1 Not lemony	2 Trace lemony	3 Faintly lemony	4 Slightly lemony	5 Moderately lemony	6 Definitely lemony	7 Strongly lemony	8 Very strongly lemony	9 Extremely lemony

Umami (Savory) Flavor

1 Not umami	2 Trace umami	3 Faintly umami	4 Slightly umami	5 Moderately umami	6 Definitely umami	7 Strongly umami	8 Very strongly umami	9 Extremely umami

Buttery Flavor

1 Not buttery	2 Trace buttery	3 Faintly buttery	4 Slightly buttery	5 Moderately buttery	6 Definitely buttery	7 Strongly buttery	8 Very strongly buttery	9 Extremely buttery

Creamy Flavor

1 Not creamy	2 Trace creamy	3 Faintly creamy	4 Slightly creamy	5 Moderately creamy	6 Definitely creamy	7 Strongly creamy	8 Very strongly creamy	9 Extremely creamy

Earthy Flavor

1 Not earthy	2 Trace earthy	3 Faintly earthy	4 Slightly earthy	5 Moderately earthy	6 Definitely earthy	7 Strongly earthy	8 Very strongly earthy	9 Extremely earthy

Remember to inhale through your mouth or nose and then exhale through your nose while chewing to maximize your flavor perception. Please be sure to rinse your palate between potato samples with the provided crackers and water.

Woody Flavor

1 Not woody	2 Trace woody	3 Faintly woody	4 Slightly woody	5 Moderately woody	6 Definitely woody	7 Strongly woody	8 Very strongly woody	9 Extremely woody

Bitter Flavor

1 Not bitter	2 Trace bitter	3 Faintly bitter	4 Slightly bitter	5 Moderately bitter	6 Definitely bitter	7 Strongly bitter	8 Very strongly bitter	9 Extremely bitter

Off-Flavors Other than Bitter

1 Not off-flavor	2 Trace off-flavor	3 Faintly off-flavor	4 Slightly off-flavor	5 Moderately off-flavor	6 Definitely off-flavor	7 Strongly off-flavor	8 Very strongly off-flavor	9 Extremely off-flavor

Mealy Texture

1 Not mealy	2 Trace mealy	3 Faintly mealy	4 Slightly mealy	5 Moderately mealy	6 Definitely mealy	7 Strongly mealy	8 Very strongly mealy	9 Extremely mealy

Overall Quality

1 Extremely low quality	2 Very strongly low quality	3 Strongly low quality	4 Definitely low quality	5 Moderate quality	6 Definitely high quality	7 Strongly high quality	8 Very strongly high quality	9 Extremely high quality