

Research Article

Characterization of the Proximate Composition, Lipid Oxidation Status, and Mineral Content of Mature Tree Nuts from Nine Hazelnut Cultivars Grown in the United States

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Hazelnuts are the most popular tree nuts in the world, and regions adjacent the Black and Mediterranean seas are the historic production centers. Characterization of hazelnut cultivars grown in these regions is well reported but is lacking for cultivars grown in the United States. The aim of our study was to characterize nine cultivars selected from the USDA National Germplasm Collection for their proximate composition, lipid oxidation status, and minerals, as well as by NIR spectroscopy. Except for ash content, proximate composition varied across the cultivars and lipids were the predominant component. NIR spectra were similar in pattern and differences in intensity could be accounted for by differences in proximate composition, including lipid, moisture, and protein. Cultivars with the highest moisture content and water activity levels were also those with highest levels of lipid oxidation. Carbon and sulfur content on a fresh weight basis varied from 44.82 g/100 g to 63.82 g/100 g and 96.56 mg/100 g to 164.79 mg/100 g, respectively. The K, P, Ca, Mg, Cu, Fe, Mn, Zn, and B contents were determined by MP-AES. Potassium followed by phosphorus was the most abundant elements. Hazelnuts appear to be a good source of dietary copper and manganese providing up 60.5% and 60.4%, respectively, of the recommended daily value while contributing no more than 0.03% of the daily value for sodium. Characterization results were in ranges like those reported for hazelnuts from Asian and European growing regions. However, each cultivar possessed a unique profile.

1. Introduction

The European hazelnut (*Corylus avellana* L.) is among the world's most popular tree nuts, second only to almonds [1]. Hazelnut is a deciduous tree cultivated in temperate climates close to large bodies of water [2]. Worldwide production has increased from 0.38 mil tons in 1970 to 1.07 mil tons by 2020, with Turkey accounting for more than 50% of the total, followed by Italy, the United States, Azerbaijan, and Chile [3].

Tree nuts are a major U.S. crop, with total production of \$9.7 billion in 2021, comprising mostly almonds (\$5.0 billion), pistachios (\$2.9 billion), and walnuts (\$1.0 billion).

California dominates the U.S. market with over 90 percent of production. At \$167 million, hazelnuts are currently a relatively minor crop exceeding only macadamia (\$63 million) in commercial production. U.S. hazelnut exports were valued at \$144 million, with top markets including Canada (\$62 million), China (\$53 million), and Mexico (\$13 million), while imports amounted to \$39 million, almost all Turkish. Historical reasons for low U.S. hazelnut production include low consumer demand and the lack of suitable cultivars for widespread commercial production.

Current domestic production is mostly in Oregon and Washington. Rising domestic and international demand for

hazelnuts along with recent successes cultivating hazelnuts in nontraditional climates (e.g., South Africa and Australia) [4] and a multiyear field trial that identified hazelnut cultivars Lewis and Ennis as potentially suitable for commercial production in California's San Joaquin Valley [5] suggests a strong potential for additional growth in U.S. production. In 2021/2022, the unit export value of \$2.22 per pound for fresh or dried hazelnuts was nearly identical to almonds or walnuts, although significantly less than pistachios (\$3.53 per pound) [6].

Given the recent extreme drought situation throughout the western United States, including California, agricultural water use is a rising concern and the source of political and cultural stress. The water footprints of almonds and pistachios are very high as compared to walnuts and hazelnuts, as much as triple depending on the data source. For instance, according to Waterfootprint.org, water consumption for almonds is 17,700 L/kg and for pistachios 12,500 L/kg compared to 5,783 L/kg for hazelnuts [7]. However, [8] distinguishes between blue water use (rivers, lakes, and reservoirs) and green water use (precipitation and groundwater) and reports blue water use (most relevant to California) of pistachios with the highest water footprints (7602 L/kg), followed by almonds (3816 L/kg), cashew nuts (3070 L/kg), walnuts (2451 L/kg), and hazelnuts (2180 L/kg). While it is apparent that water consumption by nut species has different interpretations, it is also clear that hazelnut requires substantially less than almonds or pistachios. Thus, given their popularity, comparative value, suitability for growth in California, and lower water footprint, hazelnuts seem poised to explode in future production.

Hazelnuts are consumed raw (with skin) or roasted (without skin) and used as an ingredient in a variety of foods, including chocolate, confectionery, and bakery products. Hazelnut oil is also used as a cooking oil. Shell and skin by-products from hazelnut production and processing have shown promise as feedstocks for value-added products with health-promoting properties [1]. Consumer appreciation for hazelnuts is derived from the organoleptic properties and nutritional composition of the nuts. Lipids are a chief contributor to the organoleptic properties and typically account for 50–60% of the total mass of a nut [9–11]. Phenolics are the most abundant phytochemicals in both kernels and skins, and a summary of the potential associated health benefits may be found in a review by Bottone et al. [12]. Growing evidence from clinical studies indicates that consumption of hazelnuts protects against oxidative stress and inflammation [13] and leads to lower low-density lipoprotein (LDL) and total cholesterol levels [14] without weight gain.

Hazelnuts are not self-pollinating, and orchards need several varieties of trees to produce nuts. Certain varieties produce few nuts and are planted specifically as pollinizers. Certain pollinizer varieties are compatible with certain nut-producing cultivars. Hazelnut trees in Oregon were massively infected in the 1960s by the fungal disease eastern filbert blight (EFB), and many varieties were either wiped out or significantly diminished. Researchers at Oregon State

University discovered that a particular pollinizer variety (Gasaway) possessed resistance to EFB and began a program of controlled crosses to develop resistant varieties. Since 2002, 15 of these new varieties have been released, including seven main crops and eight pollinizers [8]. Thus, hazelnut varieties are in transition, with some gone, others disappearing, and still others being created. The USDA's hazelnut germplasm collection is located in Corvallis, Oregon, and is part of larger National Plant Germplasm System (NPGS) with locations throughout the United States. NPGS is charged with the evaluation, characterization, and preservation of genetic resources. In support of this mission, nine commercially important hazelnut cultivars were selected from the collection for characterization, including proximate contents, degree of lipid oxidation, elemental analysis, and near-infrared spectroscopy (NIRS).

2. Materials and Methods

2.1. Chemicals and Reagents. Petroleum ether and hexanes (certified ACS), chloroform and methanol (HPLC grade), nitric acid (67%, Optima grade), and water ultratrace (elemental analysis grade) were purchased from Fisher Scientific Ltd. (Fair Lawn, NJ). Ferrous chloride tetrahydrate (EM Science, Darmstadt, Germany), ferric chloride hexahydrate (Fisher Scientific, Fair Lawn, NJ), and ammonium thiocyanate (LabChem Inc., Zelienople, PA) were used to determine peroxide value. Multi-element ICP calibrator IV-STOCK-8 with 24 elements at the concentration of 100 mg/L diluted in 5% nitric acid (Inorganic Ventures, Christiansburg, VA) and phosphorus of 1000 mg/L diluted in water (SPEX CertiPrep, Metuchen, NJ) were used for the preparation of standard solutions, and a custom solution of 10 elements (P, 80 mg/L; K, 200 mg/L; Ca, 50 mg/L; Mg, 40 mg/L; Na, 1.25 mg/L; Fe, 500 μ g/L; Cu, 300 μ g/L; Mn, 600 μ g/L; Zn, 300 μ g/L; and B, 300 μ g/L) diluted in 20% nitric acid (Inorganic Ventures, Christiansburg, VA) was used as a control. Water was purified and deionized to ≥ 18.1 M Ω /cm resistance using a Barnstead GenPure xCAD Plus Ultrapure Water Purification System (Thermo Scientific, Waltham, MA) and filtered through a 0.22 μ m type HA membrane filter (Millipore, Billerica, MA) before use.

2.2. Sample Preparation. Hazelnut samples harvested in 2021 were obtained from the USDA National Germplasm Repository in Corvallis OR. Upon receipt samples were stored protected from light at ambient temperature. Approximately 90 g of in-shell hazelnuts was cracked using a mortar and pestle, damaged kernels were discarded, and the remaining kernels ground using a coffee grinder. The ground material was immediately analyzed for proximate composition (ash, lipid, and moisture), water activity, and NIR spectra collected. A portion of the ground material was vacuum packed, placed at -20° C, and later sent to a commercial lab (Ward Laboratories, Inc., Kearney, NE) for %Carbon and %Nitrogen determinations.

2.3. Proximate Composition

2.3.1. Total Ash Determination. Ash content in ground hazelnut was determined using a Lindberg Blue M furnace (Thermo Fisher Scientific, Waltham, MA). 2.000 + 0.001 g of ground material was weighed in a porcelain crucible and heated on an infrared radiator in a fume hood until smoke was no longer visible. The crucibles were then placed in a furnace at 550°C for 17 h. The sample was transferred to a desiccator after heating, cooled, and weighed. Ash determination in the sample was conducted in triplicate. The ash content was calculated using the following formula:

$$\% \text{ Ash} = \frac{\text{Ash Weight (g)} \times 100}{\text{Sample Weight (g)}}. \quad (1)$$

2.3.2. Total Lipid Content. Lipids were extracted by accelerated solvent extraction using the Dionex AE350 instrument (Dionex Corp., USA). A stainless-steel extraction cell was loaded with 1.000 + 0.001 g of ground material mixed with sand and filled up to 98% of the cell capacity. The extraction was accomplished using petroleum ether at 125°C and 1500 psi for 30 min, and the extraction collected in a 60-mL amber vial. The vial was placed in a water bath at 50°C and stream of nitrogen applied for 30 min to evaporate the solvent. The resulting oil was stored at 4°C until analyses. Extraction of each sample was conducted in triplicate. Percentage of total lipid extraction was calculated by the following formula:

$$\% \text{ Total Lipid} = \frac{\text{Extracted Oil Weight (g)} \times 100}{\text{Sample Weight (g)}}. \quad (2)$$

2.3.3. Moisture Content. Moisture content was determined gravimetrically using a convection oven Model F750 (Fisher Scientific, USA). 2.000 + 0.001 g of ground material was weighed into a previously dried and weighed aluminum cup containing approximately 5.0 g of sand and a glass rod. The ground sample was dried at 105°C for 48 h and cooled in a desiccator then weighed. Moisture content in sample was determined in triplicate. Wet basis moisture was calculated by the following formula:

$$\% \text{ MC (wet)} = \frac{\text{Water Weight (g)} \times 100}{\text{Sample Weight (g)}} \quad (3)$$

2.3.4. Protein and Carbohydrate Contents. The protein content (N 6.25 for hazelnuts) was calculated using the % Nitrogen values obtained from Ward Laboratories, Inc. Total carbohydrate content was calculated by difference using the formula: $\text{total carbohydrates (g/100 g)} = 100 - (\text{g}_{\text{lipid}} + \text{g}_{\text{ash}} + \text{g}_{\text{protein}} + \text{g}_{\text{moisture}})$.

2.4. Water Activity. Water activity measurements were determined using a AquaLab 4TE (Decagon Devices, USA) and conducted in triplicate.

2.5. Lipid Oxidation. Primary and secondary oxidation was determined spectrophotometrically following the procedure of Pannico et al., 2015. In a 10-mL volumetric flask, oil extracted from 1.000 + 0.002 g of ground hazelnut was diluted with and brought up to 10 mL with hexane. The resulting solution was further diluted 1:5 with hexane, and the absorbance measured at 232, 262, 268, 270, and 274 nm using a Molecular Devices SpectraMax 384-Plus plate reader (Sunnyvale, CA). Lipid oxidation in terms of specific extinction coefficients was calculated using the following formulas:

$$K\lambda = \frac{E\lambda}{(c \times s)}, \quad (4)$$

where $K\lambda$ is the extinction coefficient at λ wavelength, $E\lambda$ is the absorbance, c is the concentration (wt/vol%), and s is the length of the cuvette (1 cm).

Lipid primary oxidation: K_{232}

Lipid secondary oxidation: K_{270}

Also, for lipid secondary oxidation:

$$\Delta K = K_{268} - \left[\frac{(K_{262} + K_{274})}{2} \right]. \quad (5)$$

Lipid oxidation for each sample was determined in triplicate from three independent extractions.

2.6. Peroxide Value. Peroxide value was analyzed following the procedure of Ribeiro [16]. In a 10-mL volumetric flask, oil extracted from 1.000 + 0.002 g of ground hazelnut was diluted and brought to 10 mL with chloroform: methanol (7:3 v/v). 2.0 mL of extract solution was transferred to a 16 × 150 mm glass disposable culture tube and combined with 7.9 mL chloroform: methanol (7:3 v/v). Then, 50 μ L of 30% (w/v) ammonium thiocyanide in water was added to the solution, followed by 50 μ L of 0.06 M ferrous chloride. The solution was mixed by vortexing after the addition of each reagent.

For the quantification of peroxide value, a ferric chloride standard curve (0–0.0150 M) was prepared from a 0.025 M ferric chloride stock solution. Briefly, 100 μ L of each calibrator was combined with 9.8 mL of chloroform-methanol solution. Then, 50 μ L of ammonium thiocyanide and 50 μ L of filtered water were added and the resulting solution mixed by vortexing. Calibrator solutions were measured before and after reading the samples.

Ferric chloride (0.0080 M) was used as a positive control, and the blank solution consisted of a 200 μ L of filtered water dissolved in 9.8 mL of the chloroform-methanol solution. The sample background was obtained by combining 9.9 mL of chloroform-methanol with 50 μ L of ammonium thiocyanide and 50 μ L of ferrous chloride. Solutions were incubated for 5 min protected from light, and the absorbance was read at 500 nm on a Molecular Devices SpectraMax 384-Plus plate reader (Sunnyvale, CA). Peroxide value was determined in triplicate using independent extracts and reported as ferric chloride milligram equivalents per gram oil.

2.7. Elemental Analysis. Inorganic residue obtained during ash determination of 2.000 g + 0.001 of ground hazelnuts was used for the analysis of macro- and microelements. Ashes contained in a porcelain crucible were dissolved in 20% nitric acid. The resulting solution was transferred to a 50-mL volumetric flask and brought up to volume. The ash acid solution was filtered through a 0.45 μm FlipMate PES/PTFE filter (Environmental Express, Charleston, SC) and collected into a 50-mL free metal centrifuge tube (Labcon, Petaluma, CA). The neat solution was used for the determination of iron, copper, manganese, boron, and sodium. The neat solution was diluted 1:4 with filtered water for zinc and phosphorus determination and the 1:4 dilution further diluted to 1:100 with 5% nitric acid for the potassium, calcium, and magnesium determinations. Analyses were conducted using an Agilent 4200 Microwave Plasma Atomic Emission Spectrophotometer (Agilent Technologies, Santa Clara, CA, USA). A custom multi-element solution (Inorganic Ventures, Christiansburg, VA) in 20% nitric acid was used as a positive control and 5% nitric acid was used as a blank. Quantification of elements was obtained using calibrators prepared from a multi-element standard solution (IV-STOCK-8, Inorganic Ventures, Christiansburg, VA) and a phosphorus standard solution (SPEX CertiPrep, Metuchen, NJ) in 5% nitric acid. Concentrations were reported in milligrams of element per a hundred grams of sample (mg/100 g sample).

2.8. NIR Measurements. Ground samples were placed in a glass vial. The mean diffuse reflectance spectra in the long-wavelength NIR regions (900 nm to 2600 nm) were measured using a benchtop FT-NIR spectrophotometer (MPA; Bruker Optics, Billerica, MA) linked to a personal computer running the OPUS software (Version 7.5; Bruker Optics, Billerica, MA). The spectrophotometer has an internal gold reference for the reflectance measurements, and the background spectrum was taken hourly. All spectra were output as absorbance.

The optical measurements were nondestructive, and the reflectance was collected from 128 individual scans in 64 cm^{-1} (every 4 to 50 nm) increments. Three measurements were taken per sample, and an average spectrum was calculated.

2.9. Statistical Analysis. Tukey's multiple comparison was used to test the difference between means of different cultivars for each organic and inorganic compound and property analyzed. Principal component analysis (PCA) was used to explore and visualize the differences between all cultivars based on all chemical analysis results.

2.9.1. NIR Spectra Pretreatment. A 5-point Savitzky-Golay first derivative and standard normal variate spectral processing were applied to all spectra to remove additive and multiplicative effects and improve the signal-to-noise ratio [17, 18]. All statistical analyses and spectral processing were performed using JMP (Pro 16; Cary, NC).

3. Results and Discussion

3.1. Proximate Contents. Proximate contents, water activity, and elemental carbon and sulfur levels found for the nine hazelnut cultivars evaluated are shown in Table 1. The highest moisture content was found in Fitzgerald (23.84%), followed by Ennis (22.07%) and Hall's Giant (16.72%). Moisture content of the remaining six cultivars was more than four times less and ranged from 6.35% (Tonda di Giffoni) to 3.09% (Giresun). Moisture contents reported in the literature span from 3.13% for Tonda Gentile Trilobata [19] to as high as 26.59% for Ordu Levant [20]. The moisture content we obtained here for Tonda di Giffoni (6.35%) was close to the 5.98% reported by [10], whereas the 3.80% measured here for Tombul was much lower than the 26.13% reported by [20]. Water activity levels in the samples ranged from 0.965 (Ennis) to 0.438 (Giresun Ordu) with those samples exhibiting the highest moisture contents also possessing the highest water activity levels. Ferrão [10] in their study of unprocessed hazelnuts from nine cultivars reported water activity levels from 0.59 to 0.80, whereas Belviso et al. [20] reported a range of 0.60 to 0.65 for dried nuts from three cultivars. Three samples in this study had water activity levels below the range reported by Ferrão et al. [10], and this was likely due to the moisture content of these samples being lower than the lowest moisture content (4.77%) measured by Ferrão et al. [10].

Ash contents for the cultivars did not show large variability and were in a narrow band ranging from 2.06% (Ennis) to 2.58% (Nixon). Ash content results obtained in the present study are within the 1.75% to 3.80% range found in the literature [9, 10, 20], and our results for Ennis (2.06%) and Hall's Giant (2.56%) are similar to those reported by Müller et al. [9] for same cultivars, 2.2% and 2.5%, respectively.

For crude protein content, Jefferson (9.21%) and Hall's Giant (17.25%) were the cultivars with the lowest and highest contents, respectively. Crude protein contents reported in the literature [9, 10, 21] ranged from 10.02% to 22.1%. In contrast to the ash results, the protein content reported by Müller et al. [9] for Ennis (12.4%) and Hall's Giant (18.4%) was slightly higher than the values we found.

Lipid content of the cultivars tested in this study ranged from 32.62% (Hall's Giant) to 65.88% (Jefferson) and is within the range of the lipid contents reported within the literature. Müller et al. [9] reported a range of 47.9% to 64.8% for 15 cultivars grown in Germany. For seven cultivars grown in Portugal [10], the lipid content ranged from 46.0% to 72.5%, and for four cultivars grown in Turkey [11], the range was 8.1% to 38.0%. Turan [20] using the cultivar Mortarella evaluated the influence of harvest year and region (2015) and canopy location (2017) on lipid content and reported ranges of 54.8% to 64.6% and 58.5.0% to 61.8%, respectively. Taken into consideration these reports and others [22 and references therein] describing lipid content over multiple cultivars, geographical regions, and harvest years, the typical lipid concentration for hazelnuts may be near to 60% but is still highly variable and dependent on multiple factors. Six of the nine cultivars evaluated fell near

TABLE 1: Proximate contents, water activity, and C and S levels determined for nine hazelnut cultivars collected from the USDA Germplasm collection.

	Moisture (%)	Ash (%)	Protein (%)	Oil (%)	Carbohydrate (%)	Water activity	Carbon (g/100 g)	Sulfur (mg/100 g)
Hall's Giant	16.72 ± 0.29	2.56 ± 0.05 ^a	17.25	32.62 ± 1.27 ^c	30.85 ± 1.31	0.922 ± 0.001	57.37	146.54
Ennis	22.08 ± 0.10	2.06 ± 0.08 ^f	11.98	36.13 ± 2.28 ^d	27.76 ± 2.28	0.965 ± 0.003 ^a	55.54	96.56
Fitzgerald	23.84 ± 0.15	2.17 ± 0.03 ^{d,e}	12.06	34.39 ± 1.44 ^{d,e}	27.53 ± 1.45	0.956 ± 0.005 ^a	44.82	124.70
Tonda di Giffoni	6.35 ± 0.09 ^a	2.40 ± 0.02 ^{b,c}	11.20	61.90 ± 2.29 ^c	18.15 ± 2.29	0.801 ± 0.003	60.01	135.68
Tombul	3.80 ± 0.09 ^{b,c}	2.34 ± 0.01 ^c	14.15	65.76 ± 1.36 ^a	13.95 ± 1.36	0.513 ± 0.001 ^b	63.06	151.05
Yamhill	4.05 ± 0.31 ^b	2.48 ± 0.03 ^{a,b}	13.36	62.88 ± 0.83 ^{b,c}	17.23 ± 0.88	0.497 ± 0.012 ^b	61.50	153.11
Nixon	6.41 ± 0.17 ^a	2.58 ± 0.03 ^a	12.43	62.66 ± 1.50 ^{b,c}	15.91 ± 1.52	0.656 ± 0.007	45.47	164.79
Jefferson	5.48 ± 0.61	2.07 ± 0.03 ^{e,f}	9.21	65.88 ± 1.96 ^a	17.37 ± 2.06	0.749 ± 0.004	57.71	109.02
Giresun	3.09 ± 0.10 ^c	2.21 ± 0.03 ^d	16.94	64.93 ± 1.23 ^{a,b}	12.83 ± 1.23	0.438 ± 0.010	63.82	159.49
Minimum	3.09	2.06	9.21	32.62	12.83	0.438	44.82	96.56
Maximum	23.84	2.58	17.25	65.88	30.85	0.965	63.82	164.79
Mean	10.20	2.32	13.18	54.13	20.17	0.722	56.59	137.88
SD	7.82	0.19	2.46	14.05	6.30	0.194	6.63	22.09

Mean reported with standard deviation for $n=3$ for moisture, ash, oil, and water activity. Standard deviation for carbohydrate calculated from standard deviation of moisture, ash, and oil. Data sharing the same letter (a, b, c, d) in the same column are not significantly different, $p > 0.05$, Tukey's multiple range tests.

this value, but those cultivars with much lower lipid contents (Hall's Giant, Fitzgerald (34.39%), and Ennis (36.13%)) were also those with the highest moisture content. Thus, it may be speculated these three cultivars may be more resistant to drying during storage or that their lipid content would have been higher had the samples been subjected to a formal drying treatment.

Carbohydrate content was calculated as the residual mass after accounting for moisture, oil, ash, and protein contents. Carbohydrate content for the samples varied from 12.83% (Giresun) to 30.85% (Hall's Giant). Carbohydrate contents reported by others have ranged from 5.3% to 22.2% [9, 10, 21]. A combination of high moisture content and lower lipid content is the reason for the elevated levels of carbohydrates in Hall's Giant (30.85%), Ennis (27.76%), and Fitzgerald (27.53%).

Total carbon and sulfur analyses are routinely performed on leafy materials to assess plant health, but results for analyses performed on nuts are not widely reported. Carbon content of the cultivars ranged from 44.82 g/100 g FW (Fitzgerald) to 63.82 g/100 g FW (Giresun). Multiple compounds, including lipids, proteins, and carbohydrates, all broadly contribute to the overall carbon content making a direct comparison to a specific class of compounds difficult, whereas most of the sulfur content may be attributed to the amino acids, methionine and cysteine, and trace-level sulfur-containing secondary metabolites [23]. For comparison purposes, the methionine and cysteine concentrations reported by Burdack-Freitag and Schieberle [23] and Alasalvar et al. [24] were used to estimate total sulfur content values of 171 mg/100 g and 121 mg/100 g, respectively. Sulfur contents determined for the samples spanned from 96.56 mg/100 g FW (Ennis) to 164.79 mg/100 g FW (Nixon) and are in the same range as the estimated values.

3.2. NIR Spectroscopy. The average raw spectra and pre-processed spectra of all nine cultivars are shown in Figures 1 and 2, respectively. The most prominent wavebands existing

in the NIR region are the strong overtone and combination absorptions of hydrogen-containing bonds O-H (found in water), C-H (found in carbohydrates and oil), and N-H (found in protein) [26]. The three cultivars, Hall's Giant, Ennis, and Fitzgerald, that have the highest moisture content and lowest lipid content can be seen separated in the processed spectra at water bands around 1500 nm and 1900 nm, as well as wavebands influenced by fats around 1400 nm, 2000 nm, and 2300 nm. Especially at 1390 nm, which is a known lipid band, the raw spectra of the three cultivars did not exhibit a peak, whereas the other six all have small peaks. Good separation at 1400 nm and 2070 nm regions also confirms with other literature that indicates these regions are the key wavelengths used for measuring lipid peroxide [27, 28]. Jefferson cultivar can be seen separated from the other cultivars at around 1700 nm and 2250 nm, which are wavebands influenced by protein [29]. This is likely due to the low protein content of Jefferson cultivar.

3.3. Lipid Oxidation Status. Peroxide values (PVs) are a measure of hydroperoxide content and are directly associated with lipid degradation. PVs in the literature range from 0.01 [19] to 7.46 meq O₂/kg oil [21]. PV results (Table 2) for the samples varied from 0.92 (Yamhill) to 7.16 (Hall's Giant) meq O₂/kg oil. Samples with the highest PVs (Hall's Giant, Ennis (6.11 meq O₂/kg oil), Fitzgerald (5.15 meq O₂/kg oil)) were also those samples with the highest moisture contents.

Spectrophotometric measurements at specific wavelengths between 232 and 274 nm are another method to assess lipid oxidation. Primary oxidation (K₂₃₂) values (Table 2) ranged from 1.25 (Tonda di Giffoni) to 1.63 (Hall's Giant), and Hall's Giant was followed by Fitzgerald (1.51) and Ennis (1.49). All the samples exhibited K₂₃₂ values lower than 2, suggesting that lipid oxidation levels were below those associated with off-flavors or rancidity [30, 31]. For secondary oxidation (K₂₇₀), the values (Table 2) ranged from 0.0901 (Giresun Ordu) to 0.2827 (Hall's Giant) and as might

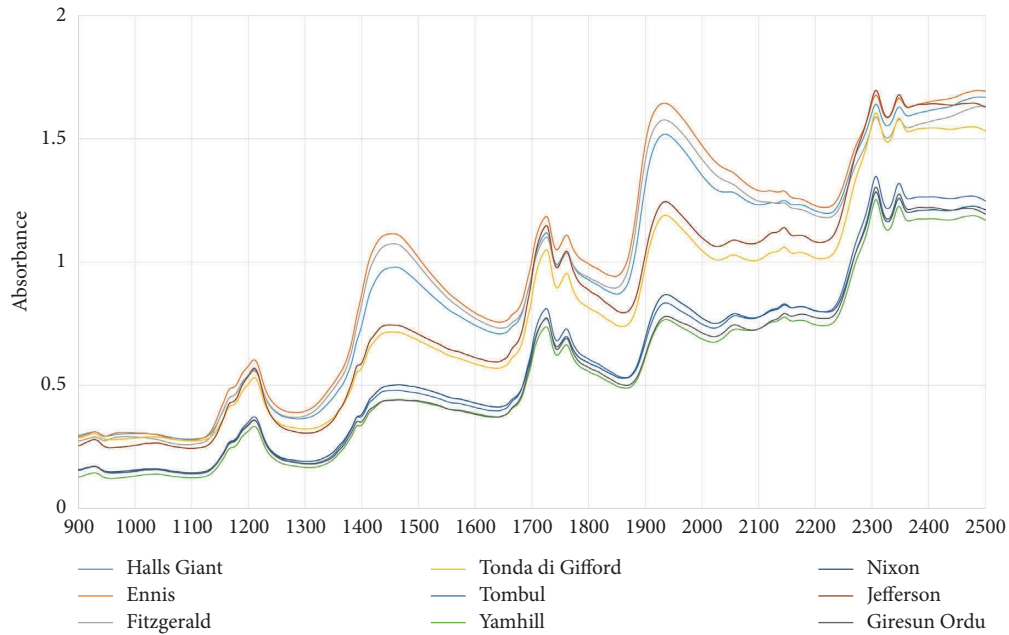


FIGURE 1: Raw spectra of nine cultivars.

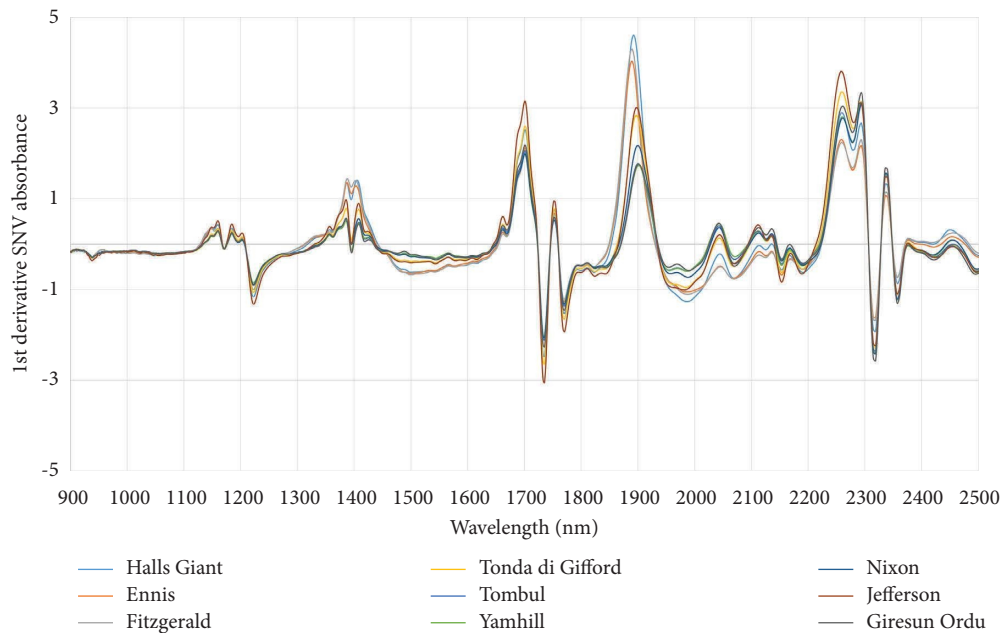


FIGURE 2: First derivative and standard normal variate preprocessed spectra of nine cultivars.

be expected Hall's Giant, Ennis (0.2499), and Fitzgerald (0.2400) also possessed the highest values for K_{270} . For ΔK (Table 2), which is another measurement of secondary oxidation, the samples with the lowest and highest values were again Giresun Ordu (0.0043) and Hall's Giant (0.0138). Primary and secondary oxidation values for the nine cultivars were comparable to the values reported in the literature [10, 21, 30].

3.4. Mineral, Macro-, and Microelements. The K, P, Ca, Mg, Cu, Fe, Mn, Zn, and B contents were determined by MP-AES using the ashed samples taken up in nitric acid, and results for Na and Mo were obtained by ICP-MS by a commercial lab (Table 3a). Mean concentrations from highest to lowest were $K > P > Mg > Ca \gg Fe > Mn > Zn > Na > Cu > B \gg Mo$. For potassium, the most abundant element of the eleven elements measured, and contents ranged from

TABLE 2: Lipid oxidation determined for nine hazelnut cultivars collected from the USDA germplasm collection.

	K323	K270	Delta K	Peroxide (mmol eq O ₂ /kg extracted oil)
Hall's Giant	1.630 ± 0.105 ^a	0.28 ± 0.02 ^a	0.014 ± 0.001	7.16 ± 0.10 ^a
Ennis	1.489 ± 0.090 ^{a,b}	0.25 ± 0.07 ^a	0.006 ± 0.000 ^{a,b,c}	6.11 ± 0.21 ^{a,b}
Fitzgerald	1.506 ± 0.053 ^{a,b}	0.24 ± 0.01 ^a	0.007 ± 0.001 ^{a,b}	5.15 ± 0.21 ^b
Tonda di Giffoni	1.255 ± 0.017 ^d	0.13 ± 0.01 ^b	0.006 ± 0.000 ^{a,b,c}	2.27 ± 0.29 ^c
Tombul	1.416 ± 0.021 ^{b,c}	0.10 ± 0.00 ^b	0.005 ± 0.000 ^{a,b,c}	2.45 ± 0.19 ^c
Yamhill	1.329 ± 0.017 ^{c,d}	0.09 ± 0.01 ^b	0.005 ± 0.001 ^{b,c}	0.92 ± 0.31 ^d
Nixon	1.322 ± 0.038 ^{c,d}	0.10 ± 0.01 ^b	0.005 ± 0.001 ^{a,b,c}	2.31 ± 0.10 ^c
Jefferson	1.277 ± 0.025 ^{c,d}	0.11 ± 0.01 ^b	0.007 ± 0.001 ^a	2.00 ± 0.08 ^{c,d}
Giresun	1.342 ± 0.014 ^{c,d}	0.09 ± 0.01 ^b	0.004 ± 0.000 ^c	1.61 ± 0.15 ^{c,d}
Minimum	1.255	0.09	0.004	0.92
Maximum	1.630	0.28	0.014	7.16
Mean	1.396	0.15	0.007	3.33
SD	0.117	0.07	0.003	2.08

Mean reported with standard deviation for $n = 3$. Data sharing the same letter (a, b, c, d) in the same column are not significantly different, $p > 0.05$, Tukey's multiple range tests.

566 mg/100 g FW (Jefferson) to 800 mg/100 g FW (Nixon) with a calculated mean of 667 mg/100 g FW. Phosphorus followed potassium with an observed range of 244 mg/100 g FW (Ennis) to 347 mg/100 g FW (Giresun) and a mean of 291 mg/100 g FW. Magnesium concentrations ranged from 139 mg/100 g FW (Ennis) to 194 mg/100 g FW (Hall's Giant) with a mean of 165 mg/100 g FW. The mean for calcium was 140 mg/100 g FW, and concentrations ranged from 97 mg/100 g FW (Fitzgerald) to 206 mg/100 g FW (Hall's Giant). For the microelement iron concentrations ranged from 2.19 mg/100 g FW (Jefferson) to 4.36 mg/100 g FW (Nixon) with a mean of 3.51 mg/100 g FW. Manganese concentrations ranged from 1.61 mg/100 g FW (Fitzgerald) to 4.90 mg/100 g FW (Hall's Giant) with a mean of 2.49 mg/100 g FW. Zinc concentrations ranged from 1.83 mg/100 g FW (Jefferson) to 3.23 mg/100 g FW (Hall's Giant) with a mean of 2.21 mg/100 g FW. Sodium concentrations ranged from 1.44 mg/100 g FW (Tonda Giffoni) to 2.24 mg/100 g FW (Nixon) with a mean of 1.72 mg/100 g FW. Boron concentrations ranged from 0.90 mg/100 g FW (Giresun) to 1.43 mg/100 g FW (Nixon) with a mean of 1.11 mg/100 g FW. For the trace element, molybdenum concentrations ranged from 1.9 µg/100 g FW (Nixon) to 25.3 µg/100 g FW (Fitzgerald) with a mean of 13.1 µg/100 g FW. There was not a single cultivar that uniformly possessed either the highest or lowest concentrations, although the cultivar Hall's Giant exhibited the highest contents of five of the elements measured (Ca, Mg, Cu, Mn, and Zn). Correlation analysis suggested that Fe contents are correlated (>0.70) with P, Ca, and Cu, that Zn contents are correlated (>0.70) with Cu and Mn, and that B contents are correlated (>0.70) with K.

Table 3c summarizes the minimum, maximum, and mean contents found in the literature for recent reports (2006–2020) on hazelnuts. The results obtained for K, P, Ca, Mg, Cu, Fe, Mn, and Zn contents for the nine cultivars tested fall within the minimum to maximum ranges, and the means observed are within one standard deviation of the values calculated from the literature results. Although not numerically large, the B, Na, and Mo contents found in some of the cultivars evaluated were lower than the values reported

in the literature, and this is more likely due to the soil and cultivation conditions than the cultivars evaluated. For example, Müller et al. [9] in their evaluation of the Mo contents of hazelnuts grown in Germany found a range of 109 µg/100 g FW to 515 µg/100 g FW, whereas Ozkutlu et al. [32] found a range of 9 µg/100 g FW to 31 µg/100 g FW for hazelnuts from the Black Sea Region of Turkey and Ozenc [33] in their evaluation of the influence of nitrogen, phosphorus, and potassium fertilizer applications on hazelnuts also from the Black Sea Region of Turkey reported a range of 40 µg/100 g FW to 51 µg/100 g FW for untreated trees and that the application of the fertilizers significantly decreased Mo contents as much as 80%.

Minerals, trace, and ultratrace elements are broadly categorized as beneficial or detrimental depending on their concentrations and biological functions. Greater than trace levels, concentrations of potassium [34], phosphorous [35], calcium [36], and magnesium [37] are required for homeostasis and good health, whereas elevated levels of sodium [38] are associated with disease. The World Health Organization has further classified Cu, Zn, and Mo as essential elements and Mn and B as probably essential elements [39] Table 3b lists the minimum and maximum percent of US FDA daily value (DV) provided by a single serving (28.35 g, 1 U.S. ounce) of hazelnuts for the nine cultivars evaluated. Our results suggest that a single serving of raw hazelnuts (28.35 g, 1 U.S. ounce) will provide 29.6% to 60.5% and 19.8% to 60.4% of the recommended DV of copper and manganese, respectively, while maintaining a low sodium diet by contributing no more than 0.03% of the DV.

3.5. Correlation among Proximate Contents, Lipid Oxidation, Mineral, Macro-, Microelements, and Principle Component Analysis (PCA). Review of Person correlations (data not shown) calculated for the properties measured revealed that moisture content was strongly correlated with peroxide value (PV), K₂₃₂, and K₂₇₀. Lipid oxidation measures (PV, K₂₃₂, K₂₇₀, and Delta K) were also strongly correlated with each other as expected. Aliasgharpour and Rahnamaye [39]

TABLE 3: (a) Fresh weight composition of minerals, trace, and ultratrace elements found in nine hazelnut cultivars collected from the USDA germplasm collection, Corvallis, Oregon. (b) US FDA recommended daily value (DV) and minimum and maximum percentages of DV provided by single servings (28.35 g) of raw hazelnuts for the nine cultivars tested. (c) Summary of minerals, trace, and ultratrace element composition found in the literature for hazelnuts adjusted to fresh weight basis.

(a)												
	K	P	Ca	Mg	Cu	Fe	Mn	Zn	B	Na	Mo	
	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(μ g/100 g)	
Hall's Giant	671 \pm 12	293 \pm 5	206 \pm 9	194 \pm 2	1.92 \pm 0.05	4.05 \pm 0.04	4.90 \pm 0.08	3.23 \pm 0.68	1.23 \pm 0.07	1.53	4.3	
Ennis	628 \pm 18	244 \pm 2	98 \pm 0.4	139 \pm 1	0.94 \pm 0.02	2.65 \pm 0.10	2.16 \pm 0.05	1.91 \pm 0.02	1.23 \pm 0.03	1.77	4.2	
Fitzgerald	687 \pm 13	259 \pm 2	97 \pm 2	140 \pm 3	1.12 \pm 0.01	3.02 \pm 0.03	1.61 \pm 0.02	2.00 \pm 0.02	0.91 \pm 0.02	1.52	25.3	
Tonda di Giffoni	734 \pm 8	304 \pm 3	134 \pm 0.4	156 \pm 1	1.86 \pm 0.03	3.57 \pm 0.01	2.98 \pm 0.03	2.08 \pm 0.03	1.28 \pm 0.01	1.44	23.1	
Tombul	651 \pm 4	299 \pm 1	160 \pm 6	164 \pm 1	1.44 \pm 0.03	3.97 \pm 0.20	1.88 \pm 0.02	2.17 \pm 0.03	1.09 \pm 0.03	1.72	16.2	
Yamhill	699 \pm 16	291 \pm 1	177 \pm 2	181 \pm 2	1.33 \pm 0.01	3.92 \pm 0.13	2.18 \pm 0.03	1.88 \pm 0.02	0.97 \pm 0.03	1.68	13.3	
Nixon	800 \pm 7	310 \pm 1	118 \pm 3	170 \pm 1	1.74 \pm 0.01	4.36 \pm 0.005	2.08 \pm 0.01	2.76 \pm 0.02	1.43 \pm 0.02	2.24	1.9	
Jefferson	566 \pm 14	268 \pm 3	98 \pm 2	188 \pm 2	1.03 \pm 0.02	2.19 \pm 0.03	2.15 \pm 0.03	1.83 \pm 0.01	0.92 \pm 0.02	1.68	12.3	
Giresun	567 \pm 2	347 \pm 1	171 \pm 1	155 \pm 1	1.34 \pm 0.01	3.82 \pm 0.01	2.43 \pm 0.001	2.00 \pm 0.02	0.90 \pm 0.01	1.90	17.2	
Minimum	566	244	97	139	0.94	2.19	1.61	1.83	0.90	1.44	1.9	
Maximum	800	347	206	194	1.92	4.36	4.90	3.23	1.43	2.24	25.3	
Mean	667	291	140	165	1.41	3.51	2.49	2.21	1.11	1.72	13.1	
SD	71	29	38	19	0.34	0.68	0.92	0.45	0.18	0.23	7.9	
(b)												
	K (mg)	P (mg)	Ca (mg)	Mg (mg)	Cu (mg)	Fe (mg)	Mn (mg)	Zn (mg)	B (mg)	Na (mg)	Mo (μ g)	
	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(mg/100 g)	(μ g/100 g)	
DV	4700	1250	1300	420	0.9	18	2.3	11	N/A	2300	45	
Minimum as % DV	3.4%	5.5%	2.1%	9.4%	29.6%	3.4%	19.8%	4.7%	—	0.02%	1.2%	
Maximum as % DV	4.8%	7.9%	4.5%	13.1%	60.5%	6.9%	60.4%	8.3%	—	0.03%	15.9%	
(c)												
N (# values)	46	46	61	61	74	77	58	77	30	31	35	
	K (mg/100 g)	P (mg/100 g)	Ca (mg/100 g)	Mg (mg/100 g)	Cu (mg/100 g)	Fe (mg/100 g)	Mn (mg/100 g)	Zn (mg/100 g)	B (mg/100 g)	Na (mg/100 g)	Mo (μ g/100 g)	
Minimum	382	192	65	54	0.76	2.36	0.68	0.29	1.36	2.04	9	
Maximum	1470	412	328	224	5.07	5.16	15.22	4.40	2.99	14.55	515	
Mean	749	282	168	145	2.21	3.72	5.61	2.68	1.98	5.30	138	
SD	186	55	47	45	0.80	0.64	3.02	0.63	0.36	3.76	157	

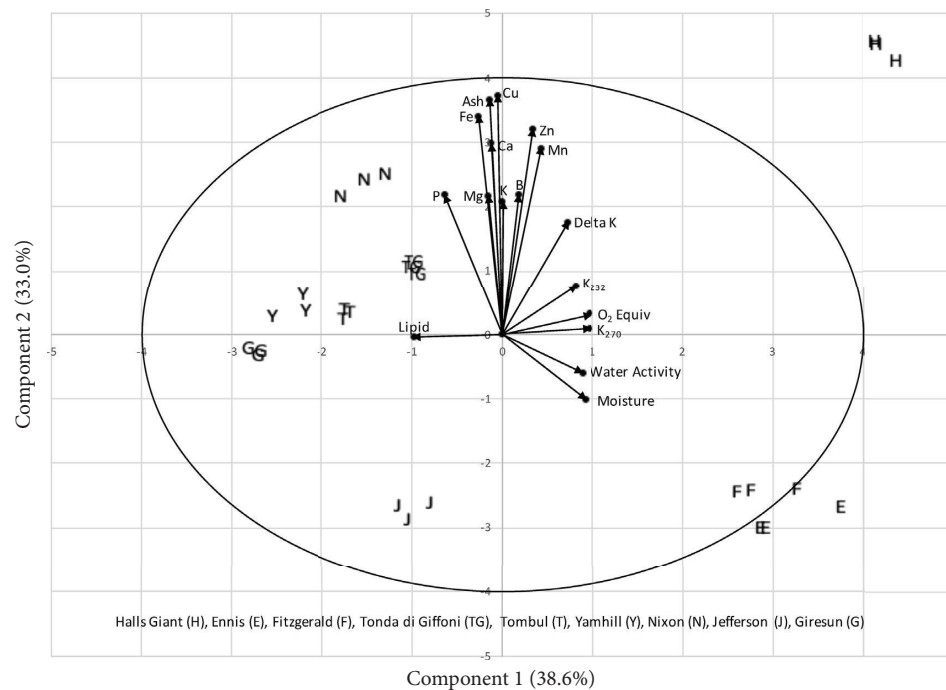


FIGURE 3: PCA Biplot showing relationships between cultivars and properties measured.

in their storability study of hazelnuts found that lipid oxidation was reduced by removing moisture, and Jung et al. [40] hypothesized that increases in lipid oxidation in samples with higher moisture content were not due to water alone, but the result of increased levels of solubilized metal ions that in turn accelerated lipid oxidation. In contrast to the positive correlations noted above, strong negative correlations were found between lipid content and both moisture and water activity, and lipid content and lipid oxidation measures (PV, K_{232} , K_{270}). Higher lipid contents leading decreased lipid oxidation may seem counterintuitive; however, this outcome might be attributable to a greater concentration of natural minor components that prevent oxidation existing in the oils [42].

Results for the PCA are displayed in Figure 3 as a biplot. One of the most apparent features is the division between the low lipid/high moisture cultivars (Hall's Giant and Ennis Fitzgerald) and the remaining cultivars. A second apparent feature is the distance between Hall's Giant and Ennis and Fitzgerald on the Component 2 axis, which can be accounted for the stark differences in ash and mineral contents between the cultivars. Likewise, similarly lower ash and mineral contents account for the separation between Jefferson and the more lipid-rich cultivars.

4. Conclusions

Hazelnuts are the most popular tree nuts in the world, and the majority of world production occurs in regions in mild climates adjacent to the large bodies of water. In the United States, almost all commercial production is limited to a single area because the cultivars used are of European and Asian descent. Only thru the introduction of new cultivars or improved

cultivation practices can U.S. and worldwide commercial production be expanded to meet the growing consumer demand accelerated by increased interest in plantbased diets and efforts to substitute nut materials for the traditional animal products in processed foods and culinary dishes [43], and overcome the pressures caused by disease and climate change [44]. The availability of germplasm collections is essential to developing new cultivars, and the NPGS contributes to these efforts through maintaining and characterizing these collections. Nine commercially important hazelnut cultivars were selected from the NPGS collection for characterization, including proximate contents, degree of lipid oxidation, elemental analysis, and near-infrared spectroscopy (NIRS). Results from the characterization demonstrated that nuts grown in the United States possess characteristics similar to hazelnuts from Asian and European growing regions, but each cultivar possessed a unique profile. Hazelnuts may contribute to nutrition as a low sodium source of lipids and dietary copper and manganese.

Data Availability

The analytical data used to support the findings of this study are included within the article.

Additional Points

Mention of a trademark or proprietary product is for identification only and does not imply a guarantee or warranty of the product by the U.S. Department of Agriculture. The U.S. Department of Agriculture prohibits discrimination in all its programs and activities on the basis of race, color, national origin, gender, religion, age, disability, political beliefs, sexual orientation, and marital or family status.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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