Comparison of Volatile Compounds Contributing to Flavor of Wild Lowbush (Vaccinium augustifolium) and Cultivated Highbush (Vaccinium corymbosum) Blueberry Fruit Using Gas Chromatography-Olfactometry

Charles F. Forney 1,*, Songshan Qiu 2, Michael A. Jordan 1, Dylan McCarthy 3 and Sherry Fillmore 1

1 Kentville Research and Development Centre, Agriculture and Agri-Food Canada, 32 Main Street, Kentville, NS B4N 1J5, Canada
2 Development Center of Technology for Fruit & Vegetable Storage and Processing Engineering, Guangdong University of Petrochemical Technology, No.139, Guandu 2nd Road, Maoming 525000, China
3 AGAT Laboratories, 11 Morris Drive, Dartmouth, NS B3B 1M2, Canada
* Correspondence: charles.forney@agr.gc.ca; Tel.: +1-902-401-8508

Abstract: The flavor of blueberry fruit products is an important parameter determining consumer satisfaction. Wild lowbush blueberries are primarily processed into products, but their flavor chemistry has not been characterized. The objective of this study was to characterize the aroma chemistry of lowbush blueberries and compare it with that of highbush. Aroma volatiles of lowbush blueberries from four Canadian provinces and five highbush blueberry cultivars were isolated using headspace solid-phase microextraction (SPME) and characterized using gas chromatography-olfactometry (GC-O) and 2-dimensional gas chromatography-time of flight-mass spectrometry (GC×GC-TOF-MS). Lowbush fruit volatiles were composed of 48% esters, 29% aldehydes and 4% monoterpenoids compared to 48% aldehydes, 26% monoterpenoids and 3% esters in highbush fruit. Twenty-three aroma-active peaks were identified in lowbush compared to forty-two in highbush fruit using GC-O. The most aroma-active compounds in lowbush fruit were ethyl 2-methylbutanoate, methyl 2-methylbutanoate, methyl 3-methylbutanoate, ethyl 3-methylbutanoate and ethyl propanoate compared to geraniol, (Z)-3-hexen-1-ol, 1-octen-3-one, α-terpineol and linalool in highbush fruit. The aroma volatile composition was more consistent among lowbush fruit samples than the five highbush cultivars. Aroma-active GC-O peaks were described more frequently as “floral”, “fruity”, “sweet” and “blueberry” in lowbush than in highbush fruit. Results suggest wild lowbush blueberries would provide “fruitier” and “sweeter” flavors to food products than cultivated highbush fruit.

Keywords: blueberry; aroma-active compounds; flavor; volatile compounds; 2-Dimensional gas chromatography-mass spectroscopy; gas chromatography-olfactometry (GC-O)

1. Introduction

The consumption of blueberry fruit and products has increased rapidly over recent years, in part due to their desirable flavor and health-promoting properties [1]. From 2018 to 2020, global blueberry production increased by over 30% [2]. Blueberry production is primarily comprised of three species of blueberries, wild lowbush (Vaccinium angustifolium Aiton), highbush (V. corymbosum L.) and rabbiteye (V. virgatum Aiton) blueberries. However, fresh and processed fruit are typically marketed without any differentiation of species.

The wild lowbush blueberry is produced commercially in northeast North America with principal production in the Canadian provinces of Nova Scotia, Prince Edward Island, New Brunswick, and Quebec and in the state of Maine [3]. The fruit is produced in managed wild stands that are composed of large numbers of different naturally occurring clones resulting in variation in fruit quality characteristics. All fruit is harvested in one harvest, and after grading, approximately 99% of the fruit is frozen for later consumption.
or processing. Fruit is distributed internationally and is used in a wide variety of food products. Wild lowbush fruit comprises over 20% of processed fruit worldwide [2].

In contrast to wild fruit, cultivated blueberry fruit are produced in plantations of named cultivars that are clonally propagated. There are many different cultivars, each having different plant and fruit characteristics. Highbush blueberry cultivars are primarily *Vaccinium corymbosum* L., but through inter-specific hybridization, the genetics of many newer cultivars include other *Vaccinium* species. This is especially true for Southern highbush blueberries where hybridization with other native *Vaccinium* species was necessary to produce cultivars adapted to warmer climates [4]. Overall, the fruit of the highbush blueberry average approximately 4 times larger than that of the wild lowbush blueberry, having more pulp, less skin and lower anthocyanins, total phenolics and antioxidant capacity than wild lowbush blueberry fruit [5]. Approximately half of the highbush blueberry fruit produced in North America are marketed as fresh fruit and half are frozen for later marketing or processing [6].

The flavor of blueberry fruit products is an important quality parameter that influences consumer satisfaction and resulting demand [7]. The chemical composition that contributes to the unique blueberry flavor includes sugars, acids and volatile compounds. Sugars are responsible for sweetness, organic acids produce tartness and volatile compounds contribute to the unique flavor and aroma of the fruit [8–11]. Blueberry aroma depends on the interaction of dozens of volatile compounds synthesized by the fruit during ripening [12]. In highbush fruit, which has been more extensively studied, approximately 120 unique volatiles have been identified [9]. Studies have shown that the volatile composition of blueberry fruit is complex and dependent on many factors including species, cultivar, environment and cultural practices [9,13,14].

Among the many blueberry volatiles reported, there has been limited determination of those responsible for blueberry aroma and flavor [9,15]. Volatile compounds that contribute to blueberry flavor have been assessed through sensory evaluation of synthetic mixtures [16,17], correlation with fruit sensory attributes [18,19] and gas chromatograph-olfactometry (GC-O) [10,20]. Partial least-squares regression correlated volatile compounds collected from highbush and rabbiteye blueberry fruit with aroma, and compounds correlated with aroma included linalool, hexanal, eucalyptol, β-caryophyllene oxide, 2-heptanone, neral, 2-undecanone and 3-methyl-1-butanol [18,19]. In Southern highbush blueberries, GC-O analysis found (E)-2-hexenal and linalool to be the most aroma-active compounds among four cultivars, while the contribution of other aroma-active compounds was cultivar-dependent and included (E,Z)-2,6-nonadienal, (Z)-3-hexenal, 2-heptanol, β-damascenone, geraniol and eugenol [10]. No identification of aroma-active compounds in wild lowbush blueberries has been reported.

Currently, blueberries are treated generically with little differentiation between species when using blueberry fruit in various food products. However, differences in fruit properties among the different commercially produced blueberry species can influence product quality attributes. To optimize the consistent flavor quality of blueberry products, a better understanding of differences in the flavor chemistry of blueberry species is needed. Therefore, the objective of this study was to identify the volatile compounds contributing to the flavor of wild lowbush blueberry fruit and compare them to cultivated highbush blueberry fruit. Increasing our understanding of differences in the flavor chemistry of blueberry fruit could improve the utilization of these fruit for the production of more flavorful blueberry products.

2. Materials and Methods

2.1. Blueberry Samples

Wild lowbush blueberry fruit were commercially harvested for the fresh market from commercial fields in Nova Scotia (NS), Prince Edward Island (PE), New Brunswick (NB) and Quebec (QC) during the 2018 season. After cleaning and grading, 500 g samples of fresh fruit were collected from three different fields in NS on 15 August; from three
different fields in PE on 31 August; from three samples taken at different times from a fresh-pack packing line in NB on 15 August; and from three different picking baskets from one field in QC on 22 August. Fruit samples were shipped overnight with ice to the Kentville Research and Development Centre (KRDC). Upon receiving, whole fruit were frozen in liquid nitrogen and stored at −80 °C until prepared for analysis. As a measure of fruit maturity, the sugar:acid ratio of the fruit was determined and averaged 14.4, 13.9, 14.0 and 15.2 for the fruit from NS, PE, NB and QC, respectively.

Cultivated highbush blueberries were hand harvested at commercial maturity from commercial fields near Centreville, Nova Scotia, Canada. Approximately 500 g of fruit from each of three fields were obtained for five cultivars ‘Duke’, ‘Brigitta’, ‘Jersey’, ‘Liberty’ and ‘Aurora’. Two harvests were obtained from each field, providing six samples for each cultivar. The genetic composition of these cultivars was 100% *V. corymbosum*, except for ‘Duke’, which was 96% *V. corymbosum* with 4% being *V. augustifolium* [4]. Harvests occurred during the 2018 season on 13 and 22 August for ‘Duke’, 15 August and 10 September for ‘Brigitta’, 20 and 27 August for ‘Jersey’, 29 August and 4 September for ‘Liberty’ and 12 and 24 September for ‘Aurora’. The day of harvest, fruit were transported to KRDC, frozen in liquid nitrogen and stored at −80 °C until prepared for analysis. The sugar:acid ratio averaged 13.4, 11.0, 10.6, 7.5 and 6.4 for ‘Duke’, ‘Brigitta’, ‘Jersey’, ‘Liberty’ and ‘Aurora’ fruit, respectively.

2.2. Gas Chromatography-Olfactometry (GC-O)

Methods used for GC-O analysis were in compliance with appropriate laws and institutional guidelines and were approved by the Agriculture and Agri-Food Canada Human Research Ethics Committee (Approval 208-F-001). Aroma-active compounds were identified using an ODP3 olfactory port (Gerstel Inc., Linthicum, MD, USA) installed on a Varian 4000 GC-MS system (Varian Inc., Walnut Creek, CA, USA). Fruit samples were taken from a −80 °C freezer and held overnight at −20 °C. A 20 g (±0.05 g) sample of fruit was combined with 80 g (±0.05 g) of a saturated NaCl solution and homogenized using a Kinematica, model MB 800 Laboratory Mixer (Kinematica AG, Luzern, Switzerland) for 1 min at a setting of 6. The blended mixture was left to settle for 10 min and then a 10 g sample was transferred to a 20 mL headspace vial that was capped with a septa lid. Vials were placed on a CombiPAL auto sampler for olfactory analysis. Prior to analysis, sample vials were held at 30 °C for 5 min, after which a divinylbenzene/carboxen/polydimethylsiloxane (DVB/CAR/PDMS) solid-phase micro extraction (SPME) fiber (Sigma-Aldrich Canada Co., Oakville, ON, Canada) was introduced into the vial headspace and allowed to adsorb headspace volatiles for 30 min. Preliminary trials found that this fiber and these adsorption conditions produced the largest amount of volatiles, which were consistent across the entire chromatogram and was similar to methods used by Du and Rouseff [10]. The SPME fiber was then desorbed for 3 min at 250 °C in the injection port of the GC onto a StabilWAX column (30 m × 0.32 mm i.d. ×1.0 μm film thickness, Restek Corporation, Bellefonte, PA, USA). The flow rate of the helium carrier gas was 2.5 mL min⁻¹ and the oven temperature was set at 50 °C for 0.2 min, then ramped at 5.0 °C min⁻¹ to 190 °C resulting in a run time of 30 min. The column effluent was split 1:1 with half going to the mass spectrometer and the other half going to the olfactory port where it was mixed with a flow of 30 mL min⁻¹ humidified air. Alkane standards (C₈-C₂₀) were run periodically and used to calculate retention index of aroma active compounds.

A trained panel of nine sensory evaluators conducted the olfactory analysis. Each replication was evaluated by five panelists. Three panelists evaluated all fruit samples, whereas the remaining six evaluators each assessed one of the three replications of each province (wild lowbush) or cultivar (cultivated highbush). Only the first harvest of the cultivated highbush fruit was subjected to olfactory analysis. Olfactory responses were collected using a touch screen and proprietary software. For each peak that was smelled, the evaluator recorded peak start time, intensity rating and olfactory descriptors by touching
virtual buttons on a touch screen. Odor intensity was rated on a scale of 1 to 5, where 1 was extremely weak, 2 was weak, 3 was moderate, 4 was strong and 5 was extremely strong. Evaluators described the odor smelled by selecting one or more of sixteen descriptors that had been predetermined by the panel based on preliminary training sessions. The sixteen descriptors were “fruity”, “blueberry”, “citrus”, “floral”, “green-grassy”, “herb-like”, “sweet”, “caramel”, “roasted-nutty”, “earthy-musty”, “rancid-cheesy”, “sulfury”, “acidic-vinegar”, “pungent-sharp”, “chemical” and “other”. In addition, evaluators could note additional descriptors by recording the peak start time and descriptor on a note pad. The intensity and frequency of each aroma-active peak was summarized by calculating the modified frequency (MF) values, which provided an overall measure of the compound’s contribution to aroma [21–24]. The MF values were calculated using the formula $MF(\%) = \left(\frac{F(\%)}{I(\%)}\right)^2$, where $F(\%)$ was the frequency of odor detection among evaluators as a % of all evaluators, and $I(\%)$ was the average intensity as a percentage of the maximum intensity (5). Aroma peaks with MF values < 25% were not considered significant contributors to blueberry aroma. The identification of aroma-active peaks was determined by matching RI values and aroma descriptions with that of pure standards, as well as peak identification by two-dimensional gas chromatography–time of flight–mass spectroscopy (GC×GC-TOF-MS).

2.3. 2Dimensional Gas Chromatography-Time of Flight-Mass Spectrometry

To aid in the identification of aroma-active compounds and determine volatile profiles for wild lowbush and cultivated highbush blueberry fruit, samples were subjected to GC×GC-TOF-MS. Headspace volatile samples were collected from homogenized fruit samples that were prepared as described above. Blanks were made by transferring 10 g of the saturated salt solution into a headspace vial. In addition, retention index standards were prepared by injecting 5 µL of 10 µg µL$^{-1}$ C$_8$-C$_{20}$ alkanes into a 20 mL headspace vial. All prepared vials were immediately placed in the autosampler rack of a MultiPurpose Sampler (MPS) (Gerstel, Linthicum, MD, USA) for analysis on a Pegasus 4D GC×GC-TOF-MS (LECO, St. Joseph, MI, USA). Sample vials were then held at 30 °C for 5 min, after which a DVB/CAR/PDMS SPME fiber (Sigma-Aldrich Canada Co., Oakville, ON, Canada) was introduced into the vial headspace and allowed to adsorb headspace volatiles for 30 min. The SPME fiber was then desorbed for 3 min at 250 °C in the injection port of the GC, followed by 4 min of conditioning at 250 °C. Helium was used as the carrier gas, and the injector operated with a 1:10 split for 1 min following the introduction of the SPME fiber. This split ratio was chosen to maximize detection sensitivity while preventing saturation of the detector. The column flow was maintained at 1.4 mL min$^{-1}$. The GC×GC system had a polar StabilWAX column (30 m × 0.25 mm i.d. × 0.25 µm film thickness, Restek Corporation, Bellefonte, PA, USA) for the first dimension and a mid-polar RXI-5Sil column (0.6 m × 0.25 mm i.d. × 0.25 µm film thickness, Restek Corporation, Bellefonte, PA, USA) for the second dimension. The two columns were interfaced with a liquid-nitrogen-cooled dual-stage cryogenic modulator and the second column was located in an oven with the temperature program independent of the first-dimension column oven. The GC×GC operating conditions were optimized using Simply GC×GC™ (LECO, St. Joseph, MI, USA). The temperature program for the primary GC oven was set at 50 °C for 0.2 min, then ramped at 10.3 °C min$^{-1}$ to 220 °C. The secondary oven was maintained 33 °C warmer than the primary oven. The modulation period, the hot-pulse duration and the cooling time between stages were set at 1.3, 0.39 and 0.26 s, respectively. The transfer line to the TOF-MS detector source was maintained at 250 °C. The ion source temperature was 250 °C with a filament voltage of 70 eV. The data acquisition rate was 200 spectra s$^{-1}$ for the mass range of 35–300 amu. Mass calibration and tuning were conducted daily using perfluorotributylamine (PFTBA).

Compound identification was based on the retention index (RI) and similarity with the National Institute of Standards and Technology (NIST) Mass Spectral Virtual Library (ChemSW, Fairfield, CA, USA). Identifications were also confirmed using known
standards when available. Data were processed using LECO ChromaTOF software (LECO, St. Joseph, MI, USA), and an estimate of the peak area counts of each compound was calculated using the LECO APEX data deconvolution/processing routine.

To aid in the identification of aroma-active compounds that were present in low concentrations, additional analyses were conducted. To increase the sensitivity of detection, 20 g of fruit tissue was homogenized in 30 g of a saturated NaCl solution and 10 mL of this homogenate was transferred to a 20 mL headspace vial. Analysis of the headspace volatiles was conducted as described above, except the injector operated with a 1:5 split for 1 min following the introduction of the SPME fiber to the GC injection port.

2.4. Statistical Analysis

The volatile data for the wild lowbush fruit were collected from a designed experiment with random effects of three replicates from four different provinces, and the fixed effect was the differences between the provinces. The volatile data for the cultivated highbush fruit had three fields from five different cultivars for the random effects and the fixed effects were the cultivars. The volatile data were analyzed by ANOVA using the statistical software Genstat 16 (VSN International, Hemel Hempstead, England, UK). Differences were considered to be significant at $p < 0.05$. For analysis of the frequency of aroma descriptors, GC-O aroma peaks were restricted to those that were identified by two or more of the five evaluators that assessed each fruit sample. The total number of each descriptor for each sample was analyzed by ANOVA, and differences among provinces and cultivars and between wild lowbush and cultivated highbush blueberries were determined by the least significant difference test (LSD$_{0.05}$). To further explore differences in the aroma profiles of the wild lowbush and cultivated highbush blueberry fruit, volatiles were totaled via chemical classification, and principal component analysis using correlations of Euclidian distances was conducted using Genstat 16.

3. Results and Discussion

3.1. Aroma-Active Volatiles of Wild Lowbush Blueberries

Twenty-three aroma-active peaks that had average MF values $> 25\%$ were identified in wild lowbush blueberries using GC-O and represented twenty-five compounds (Table 1). Of these, eight were identified as esters, five alcohols, five ketones, four aldehydes, two terpenes and one unknown. Most aroma-active compounds were found in fruit from all four provinces at similar MF values. All twenty-three peaks were detected in fruit from NS and QC. Geraniol and 3-heptanone were not detected in fruit from PE and ($E$)-2-nonenal was not detected in fruit from NB. In addition, ethyl butanoate was only detected by GC-O in fruit from QC and had an MF value of 32.7\% (data not shown). These differences may reflect genetic diversity in the wild blueberry fields from the different provinces [5]. Furthermore, differences in fruit maturity, environmental growing conditions in the different provinces and/or handling could impact aroma-active volatile synthesis and composition, which has been reported in rabbiteye and highbush blueberries [14,15,25].

Through GC-O analysis, the aroma-active peaks with the greatest MF values in wild lowbush blueberry fruit were identified as the esters ethyl 2-methylbutanoate, methyl 2-methylbutanoate, methyl 3-methylbutanoate, ethyl 3-methylbutanoate and ethyl propanoate, all having average MF values $> 50\%$ (Table 1). These compounds were described as “fruity” and “sweet” with three being described as “blueberry”. The four branched-chain esters were abundant in wild lowbush blueberry fruit and comprised over 29\% of the total volatile compounds, while the straight-chain ester ethyl propanoate only accounted for 0.13\% (Table 2). All of these esters were previously reported among headspace volatiles collected from whole lowbush blueberry fruit except for methyl 2-methylbutanoate [26]. 3-Hexanone, which was found in low concentrations, coeluted with ethyl 2-methylbutanoate and may have contributed to the “sweet” and “fruity” aroma of this peak. Additional esters detected in this study by GC-O and contributing “fruity” and “sweet” aromas included
methyl 2-pentenoate, ethyl 2-methylpropanoate and methyl 3-methyl-2-butenoate. Lugemwa et al. [26] also reported these esters in lowbush blueberry fruit.

Table 1. Aroma-active compounds in wild lowbush blueberry fruit from the provinces of Nova Scotia (NS), Prince Edward Island (PE), New Brunswick (NB) and Quebec (QC) identified through gas chromatography-olfactometry (GC-O) and ranked by average modified frequency (MF) values.

| Compound                                      | RI     | MF Value (%) | Aroma Descriptors 2 | ID Basis 3     |
|-----------------------------------------------|--------|--------------|---------------------|----------------|
| Ethyl 2-methylbutanoate/3-Hexanone            | 1052   | 70.6         | 74.0                | Fruity (30), Sweet (30), Blueberry (17), Floral (10), Citrus (4) |
| Methyl 2-methylbutanoate                      | 1012   | 72.1         | 67.1                | Sweet (32), Fruity (21), Blueberry (15), Floral (12), Caramel (8) |
| Methyl 3-methylbutanoate                      | 1022   | 64.5         | 60.1                | Fruity (15), Sweet (14), Rancid-cheesy (12), Pungent-sharp (10), Blueberry (8) |
| Ethyl 3-methylbutanoate                       | 1066   | 48.1         | 54.2                | Fruity (19), Sweet (18), Blueberry (11), Floral (9), Herb-like (3), Pungent-sharp (3), Rancid-cheesy (3) |
| Ethyl propanoate                              | 951    | 53.7         | 55.1                | Citrus (13), Green-grassy (11), Sweet (8), Chemical (6), Fruity (6), Floral (6) |
| (E)-2-Hexen-1-ol                              | 1401   | 57.5         | 44.7                | Earthy-musty (9), Pungent-sharp (8), Rancid-cheesy (19), Sulfury (13) |
| (Z)-3-Hexen-1-ol                              | 1378   | 52.5         | 42.3                | Green-grassy (18), Rancid-cheesy (10), Earthy-musty (9), Herb-like (8), Sweet (5) |
| 1-Octen-3-one/Methyl 2-pentenoate 5           | 1303   | 64.5         | 49.9                | Earthy-musty (18), Rancid-cheesy (9), Herb-like (9), Pungent-sharp (8), Rancid-cheesy (5) |
| 2-Dodecanone                                  | 1719   | 48.4         | 48.1                | Floral (16), Fruity (10), Herb-like (10), Citrus (10), Sweet (9) |
| Ethyl 2-methylpropanoate                      | 970    | 51.4         | 49.9                | Sweet (23), Fruity (15), Floral (8), Citrus (6), Blueberry (4) |
| Linalool                                      | 1531   | 51.4         | 46.5                | Floral (22), Sweet (19), Fruity (11), Herb-like (6), Blueberry (6) |
| Geraniol                                      | 1827   | 63.2         | 65.4                | Floral (18), Sweet (10), Citrus (6), Fruity (5), Blueberry (5) |
| 2-Ethyl-1-hexanol                             | 1504   | 44.2         | 46.2                | Sweet (11), Floral (10), Herb-like (10), Citrus (9), Green-grassy (8) |
| Unknown 786                                   | 786    | 40.0         | 39.0                | Rancid-cheesy (18), Earthy-musty (8), Pungent-sharp (4), Blueberry (3), Floral (3), Herb-like (3), Floral (10), Sweet (7), Fruity (5) |
| (E)-2-Hexenal                                 | 1226   | 20.7         | 51.4                | Green-grassy (4), Herb-like (4), Roasted-nutty (4) |
| 1-Penten-3-one                                | 1031   | 25.3         | 46.2                | Rancid-cheesy (9), Earthy-musty (5), Floral (3), Green-grass (3), Herb-like (3), Pungent-sharp (3) |
| Hexanal                                       | 1087   | 32.7         | 48.1                | Green-grassy (20), Herb-like (6), Blueberry (2), Earthy-musty (2), Roasted-nutty (2), Fruity (2) |
| 1-Pentanol                                    | 1245   | 32.7         | 47.1                | Citrus (4), Roasted-nutty (3), Fruity (3), Rancid-cheesy (3), Pungent-sharp (3), Floral (3) |
| Methyl 3-methyl-2-butenoate                   | 1175   | 38.6         | 37.9                | Sweet (12), Fruity (11), Herb-like (5), Blueberry (3), Floral (3), Caramel (3) |
| 1-Hexanol                                     | 1345   | 33.5         | 37.4                | Fruity (11), Sweet (10), Blueberry (4), Blueberry (4), Caramel (2) |
| (Z)-3-Hexenal                                 | 1149   | 25.8         | 30.6                | Green-grassy (13), Herb-like (6), Rancid-cheesy (4), Fruity (3), Floral (3) |
| 3-Heptanone                                   | 1152   | 23.1         | 35.8                | Green-grassy (14), Earthy-musty (7), Floral (4), Herb-like (3), Fruity (2), Roasted-nutty (2) |
| (E)-2-Nonenal                                  | 1541   | 42.2         | 36.6                | Earthy-musty (8), Roasted-nutty (6), Rancid-cheesy (5), Sweet (4), Fruity (3) |

1 Values for each province are the average of fifteen GC-O analyses conducted on fruit from three commercial lowbush fields by five evaluators for each sample. Compounds presented had average MF values > 25%. 2 The five most frequent descriptors. 3 RI, retention index; Std, standard; AD aroma description; MS mass spectrum (See Table 2 for RI reference comparisons and MS similarity values). 4 Frequency of descriptor chosen by GC-O panelists. 5 Tentative identification.
Table 2. Headspace volatile composition of wild lowbush blueberry fruit from the provinces of Nova Scotia (NS), Prince Edward Island (PE), New Brunswick (NB) and Quebec (QC) determined by 2-dimensional gas chromatography–time of flight–mass spectrometry (GCxGC-TOF-MS) ¹.

| Compound               | RI     | RI-Ref | Similarity ³ | Volatile Composition (Area Counts) | F Prob ⁴ | % |
|------------------------|--------|--------|--------------|-----------------------------------|----------|---|
|                        |        |        |              | NS       | PE       | NB       | QC       | Grand Mean | SEM      |    |
| Acids                  |        |        |              |          |          |          |          |           |          |    |
| Octanoic acid          | 2059   | 2051   | 902.2        | 4429     | - ⁵      | 6898     | 8099     | 4857       | 5916     | ns 0.93 |
| 2-Ethylhexanoic acid   | 1946   | 1934   | 926.0        | 3587     | -        | 2396     | 4468     | 2613       | 2962     | ns 0.50 |
| Heptanoic acid         | 1952   | 1953   | 914.5        | 2383     | -        | 2442     | 3380     | 2051       | 2390     | ns 0.39 |
| R-4-Methylhexanoic acid| 1928   | 1925   | 852.0        | 1096     | -        | 981      | 1512     | 897        | 1042     | ns 0.17 |
| 5-Methylhexanoic acid  | 1907   | 1914   | 910.3        | 825      | -        | 734      | 1204     | 691        | 812      | ns 0.13 |
| 3-Methylhexanoic acid  | 1886   | 1869   | 894.0        | 563      | -        | 651      | 962      | 544        | 657      | ns 0.10 |
| Total acids            | 12,884 | 0.000  | 14,102       | 19,625   | 11,653   | 13,779   |           |           |          | 2.24    |
| Alcohols               |        |        |              |          |          |          |          |           |          |    |
| 2-Ethyl-1-hexanol      | 1486   | 1491   | 934.0        | 84,188   | 961      | 12,129   | 10,615   | 26,973     | 24,867   | ns 5.17 |
| (E)-2-Hexen-1-ol       | 1404   | 1402   | 935.0        | 3385     | 4479     | 4458     | 9004     | 5332       | 1241     | 0.068 1.02 |
| 1-Hexanol              | 1349   | 1370   | 902.6        | 2966     | 1915     | 1900     | 7792     | 3643       | 973      | 0.015 0.70 |
| Ethanol                | 929    | 929    | 944.0        | 2140     | 390      | 1335     | 9875     | 3435       | 2702     | ns 0.66 |
| 1-Pentanol             | 1245   | 1241   | 895.0        | 1715     | 2169     | -        | 3920     | 1951       | 1591     | ns 0.37 |
| 3-Methyl-1-butanol     | 1202   | 1204   | 913.0        | 326      | 351      | 1716     | 4004     | 1599       | 651      | 0.022 0.31 |
| (Z)-3-Hexen-1-ol       | 1382   | 1381   | 932.0        | 906      | 817      | 698      | 3227     | 1412       | 411      | 0.013 0.27 |
| 2-Hexyn-1-ol           | 1205   | 1207   | 885.5        | 1355     | 1477     | 827      | 267      | 981        | 721      | ns 0.19 |
| 1-Heptanol             | 1451   | 1463   | 893.0        | 781      | -        | 182      | 195      | 289        | 256      | ns 0.06 |
| Total alcohols         | 97,762 | 12,559 | 23,244       | 48,899   | 45,616   | 33,412   |           |           |          | 8.75    |
| Aldehydes              |        |        |              |          |          |          |          |           |          |    |
| (E)-2-Hexenal          | 1223   | 1216   | 940.0        | 27,315   | 101,405  | 43,858   | 38,607   | 52,796     | 15,783   | 0.058 10.13 |
| Hexanal                | 1080   | 1087   | 911.0        | 53,158   | 40,319   | 27,297   | 24,565   | 36,335     | 14,383   | ns 6.97 |
| (Z)-3-Hexenal          | 1144   | 1142   | 870.0        | 10,064   | 34,454   | 30,491   | 19,731   | 23,685     | 13,809   | ns 4.54 |
| Heptanal               | 1186   | 1182   | 892.5        | 43,871   | 891      | 6925     | 4602     | 4072       | 14,755   | ns 2.76 |
| 2-Ethylhexanal         | 1188   | 1210   | 935.0        | 32,448   | -        | 887      | 789      | 8531       | 11,343   | ns 1.64 |
| Pentanal               | 982    | 981    | 897.0        | 9001     | 2580     | 3061     | 6614     | 5314       | 2160     | ns 1.02 |
| (E,E)-2,4-Hexadienal   | 1405   | 1414   | 924.9        | 1043     | 2489     | 1979     | 1472     | 1736       | 746      | ns 0.33 |
| Octanal                | 1291   | 1288   | 920.0        | 3339     | 524      | 686      | 487      | 1289       | 1065     | ns 0.24 |
| Nonanal                | 1396   | 1392   | 883.0        | 1281     | 996      | 1223     | 1123     | 1156       | 477      | ns 0.22 |
| (E)-2-Heptanal         | 1328   | 1319   | 915.0        | 1552     | 1038     | 505      | 965      | 1015       | 504      | ns 0.19 |
| 3-Methylbutanal        | 918    | 912    | 868.0        | 1286     | 610      | -        | 775      | 668        | 381      | ns 0.13 |
| 3-Methylpentanal       | 1034   | 1032   | 829.6        | 2212     | -        | 408      | -        | 655        | 595      | ns 0.13 |
| 2-Pentenal             | 1133   | 1073   | 860.6        | 656      | 382      | 148      | 499      | 421        | 396      | ns 0.08 |
| 2-Methylpentanal       | 966    | na     | 862.6        | 1135     | -        | 243      | -        | 343        | 356      | 0.07 |
| 4-Methylhexanal        | 1158   | na     | 815.0        | 1233     | -        | -        | 308      | 478        | 0.06     |
| Methacrolein           | 888    | 886.3  | 913.0        | 965      | 216      | -        | -        | 295        | 323      | 0.06 |
| Unknown 1165           | 1165   | NA     | 845.0        | 1041     | -        | -        | -        | 260        | 521      | 0.05 |
| Total                  | 191,601| 185,863| 117,712      | 100,229  | 148,851  | 78,165   |           |           |          | 28.55  |
Table 2. Cont.

| Compound                      | RI   | RI-Ref | Similarity | Volatile Composition (Area Counts) | F Prob | %   |
|-------------------------------|------|--------|------------|------------------------------------|--------|-----|
|                               |      |        |            | NS | PE | NB | QC | Grand Mean | SEM |
| Amines                        |      |        |            |    |    |    |    |             |     |
| Dimethylamine                 | 882  | na     | 893.5      |    |    |    |    | 1497 | 1334 | 708 | 1083 | ns | 0.14 |
| Total                         |      |        |            |    |    |    |    | 1497 | 1334 | 708 | 1083 | ns | 0.14 |
| Esters-Branched Chain         |      |        |            |    |    |    |    |      |      |     |     |    |     |
| Methyl 3-methylbutanoate      | 1017 | 1018   | 943.5      | 76446 | 102763 | 125131 | 95571 | 99978 | 28679 | ns | 19.18 |
| Ethyl 3-methylbutanoate       | 1063 | 1068   | 948.3      | 13835 | 13575 | 28727 | 83042 | 34795 | 15906 | 0.061 | 6.67 |
| Methyl 2-methylbutanoate      | 1012 | 1048   | 901.9      | 10061 | 9057 | 24997 | 13592 | 14427 | 4852 | ns | 2.77 |
| Ethyl 2-methylbutanoate       | 1046 | 1052   | 939.8      | 3362 | 2575 | 6763 | 12207 | 6227 | 3146 | ns | 1.19 |
| Methyl 3-methyl-2-butenoate   | 1170 | 1170   | 937.0      | 1806 | 2588 | 3925 | 1022 | 2278 | 746 | ns | 0.44 |
| Methyl 3-methyl-3-butenoate   | 1118 | na     | 926.0      | 1855 | 1961 | 2993 | 407 | 1704 | 306 | 0.012 | 0.33 |
| Methyl 2-methylpropanoate     | 921  | 919    | 889.8      | 600 | 778 | 3579 | 969 | 1481 | 366 | 0.004 | 0.28 |
| 3-Methylbutyl acetate         | 1120 | 1124   | 884.7      | 326 | 392 | 4359 | 99 | 1294 | 556 | 0.004 | 0.25 |
| Methyl 3-hydroxy-3-methylbutanoate | 1375 | 1374   | 884.3      | 523 | 2636 | 1245 | 271 | 1169 | 1269 | ns | 0.22 |
| Ethyl 2-hydroxy-3-methylbutanoate | 1426 | 1422   | 922.0      | 904 | 922 | 1661 | 514 | 770 | 403 | ns | 0.15 |
| Ethyl 2-methylenepropanoate   | 961  | 960    | 918.0      | 127 | 1473 | 1170 | 693 | 375 | 0.074 | 0.13 |
| Total                         |      |        |            | 109844 | 136094 | 204453 | 208864 | 164814 | 56604 | ns | 31.62 |
| Esters-Straight Chain         |      |        |            |    |    |    |    |      |      |     |     |    |     |
| Ethyl Acetate                 | 888  | 879    | 953.0      | 65690 | 10775 | 89966 | 67634 | 58516 | 13283 | 0.026 | 11.23 |
| Methyl acetate                | 825  | 815    | 973.7      | 15204 | 3424 | 29942 | 22563 | 17783 | 5228 | 0.052 | 3.41 |
| Methyl butanoate              | 986  | 983    | 942.0      | 1382 | 1173 | 2380 | 2994 | 1982 | 1007 | ns | 0.38 |
| Ethyl butanoate               | 1032 | 1032   | 894.5      | –   | –   | 919 | 3931 | 1212 | 1564 | ns | 0.23 |
| (E)-Hexyl acetate             | 1333 | 1334   | 888.6      | 212 | 857 | 935 | 1477 | 870 | 620 | ns | 0.17 |
| Ethyl propanoate              | 954  | 952    | 874.6      | 289 | 0   | 1338 | 1034 | 665 | 617 | ns | 0.13 |
| Methyl propanoate             | 907  | 906    | 937.5      | 401 | 422 | 1561 | 221 | 651 | 264 | 0.039 | 0.12 |
| Pentyl acetate                | 1120 | 1175   | 875.5      | –   | –   | –   | 2511 | 628 | 891 | ns | 0.12 |
| Methyl 2-pentenoate           | 1350 | na     | 729.4      | 688 | 698 | 855 | –   | 560 | 569 | ns | 0.11 |
| Total                         |      |        |            | 83866 | 17349 | 127897 | 102364 | 82869 | 24042 | 0.016 | 15.90 |
| Furans                        |      |        |            |    |    |    |    |      |      |     |     |    |     |
| 2-Ethylfuran                  | 954  | 949    | 875.0      | 558 | 511 | 529 | 411 | 502 | 421 | ns | 0.10 |
| 2-Ethyl-5-methylenehydrofuran | 939  | na     | 777.6      | 228 | 151 | 529 | 856 | 309 | 428 | ns | 0.06 |
| Total                         |      |        |            | 786 | 663 | 529 | 1267 | 811 | 849 | ns | 0.16 |
| Hydrocarbons                  |      |        |            |    |    |    |    |      |      |     |     |    |     |
| Toluene                       | 1041 | 1042   | 912.5      | 9754 | 1147 | –   | 2228 | 3282 | 2965 | ns | 0.63 |
| (Z)-1-Ethyl-2-methylcyclopropane | 1061 | 1062   | 752.5      | –   | –   | 7323 | 409 | 1933 | 3701 | ns | 0.37 |
| Benzene                       | 946  | 943    | 965.0      | 7207 | –   | 206 | 0 | 1853 | 1872 | 0.084 | 0.36 |
| Hexane                        | 600  | 600    | 909.0      | 1085 | 1138 | 1729 | 783 | 1183 | 380 | ns | 0.23 |
| m-Xylene                      | 1144 | 1144   | 953.6      | 1002 | 2253 | –   | –   | 814 | 322 | 0.007 | 0.16 |
| Ethylbenzene                  | 1130 | 1141   | 916.4      | 2037 | 826 | –   | –   | 716 | 534 | ns | 0.14 |
Table 2. Cont.

| Compound                          | RI  | RI-Ref | Similarity | Volatile Composition (Area Counts) | F Prob | %  |
|-----------------------------------|-----|--------|------------|--------------------------------------|--------|----|
|                                  |     | RI-Ref |            | NS        | PE   | NB   | QC   | Grand Mean | SEM   |     |
| 4-Methyl-1,3-pentadiene           | 782 | 796    | 946.8      | 227       | 470  | 876  | 757  | 583        | 260   | ns  | 0.11 |
| o-Xylene                          | 1190| 1188   | 934        | 984       | 975  | –    | –    | –          | 490   | 511 | 0.09 |
| 2-Octene                          | 841 | 858    | 861.8      | 1217      | –    | –    | –    | 220        | 359   | 396 | 0.07 |
| Indene                            | 1495| 1471   | 927.0      | 1117      | –    | –    | –    | –          | 279   | 282 | 0.073 | 0.05 |
| 4,4-Dimethyl-1,2-pentadiene       | 954 | na     | 721.7      | 556       | 357  | –    | 108  | 255        | 370   | ns  | 0.05 |
| 3-Propoxy-1-propene               | 1007| na     | 865.0      | 938       | –    | –    | –    | 235        | 469   | ns  | 0.04 |
| Total                             |     |        |            | 26,124    | 7165 | 10,134| 4505 | 11,982     | 12,063| ns  | 2.30 |
| Ketones                           |     |        |            |           |      |      |      |            |       |     |      |
| 2-Heptanone                       | 1182| 1184   | 922.5      | 19,191    | 8673 | 6086 | 4419 | 9592       | 6738  | ns  | 1.84 |
| 2-Nonanone                        | 1389| 1390   | 909.75     | 3152      | 6738 | 2002 | 7649 | 4885       | 2166  | ns  | 0.94 |
| 3-Heptanone                       | 1152| 1151   | 865        | 9414      | 231  | 2065 | 616  | 3081       | 2745  | ns  | 0.59 |
| 4-Heptanone                       | 1124| 1142   | 912.75     | 6389      | –    | 1833 | 176  | 2100       | 1874  | ns  | 0.4 |
| 5-Methyl-3-methylene-2-hexanone   | 1254| na     | 811.0      | 8056      | –    | –    | 2014 | 3120       | ns    | 0.39 |
| 6-Methyl-5-hepten-2-one           | 1339| 1340   | 897.9      | 1633      | 1768 | 1165 | 2348 | 1729       | 409   | ns  | 0.33 |
| 2-Methyl-2-heptan-4-one           | 1214| NA     | 808.5      | 5568      | –    | –    | –    | 1392       | 2260  | ns  | 0.27 |
| 3-Hexanone                        | 1049| 1047   | 898        | 4281      | –    | 226  | –    | 1127       | 1325  | ns  | 0.22 |
| 2-Butanone                        | 904 | 903    | 843.7      | 4053      | 159  | –    | –    | 1035       | 1070  | 0.09 | 0.2 |
| 3-Methyl-3-buten-2-one            | 996 | 996    | 922.8      | 616       | 1265 | –    | 1229 | 778        | 726   | ns  | 0.15 |
| 1-Penten-3-one                    | 1052| 1030   | 881.75     | 664       | 826  | 186  | 1011 | 672        | 520   | ns  | 0.13 |
| 2-Methylcyclopentanone            | 1198| 1187   | 841.0      | 2125      | –    | –    | –    | 531        | 703   | ns  | 0.1 |
| 5-Hexen-2-one                     | 1130| 1137   | 939.0      | 1673      | –    | –    | –    | 418        | 564   | ns  | 0.08 |
| Acetone                           | 819 | 817    | 958.5      | 1110      | 190  | –    | 256  | 389        | 319   | ns  | 0.07 |
| 2-Octanone                        | 1286| 1290   | 915.0      | 1499      | –    | –    | –    | 375        | 483   | ns  | 0.07 |
| 1-Octen-3-one                     | 1304| 1301   | 842.5      | 1446      | –    | –    | 361  | 531        | ns    | 0.07 |
| 3-Methyl-2-butanone               | 932 | 939    | 826.0      | 1359      | –    | –    | –    | 340        | 528   | ns  | 0.07 |
| 3-Hexen-2-one                     | 1218| 1212   | 903.0      | –         | –    | –    | 117  | 29         | 58    | ns  | 0.01 |
| Total                             |     |        |            | 72,230    | 19,851| 13,564| 17,822| 30,866     | 26,137| ns  | 5.92 |
| Monoterpenoids                    |     |        |            |           |      |      |      |            |       |     |      |
| Linalool                          | 1542| 1540   | 897.4      | 10,974    | 8268 | 12,813| 13,949| 11,501     | 3640  | ns  | 2.21 |
| (2R,5S)-2-Methyl-5-(prop-1-en-2-yl)-2-vinyltetrahydrofuran | 1243| 1226 | 887 | 2217 | 2380 | 4403 | 3021 | 3005 | 1601 | 0.58 |
| α-Terpineol                       | 1698| 1690   | 922.5      | 1589      | 924  | 1739 | 2598 | 1713       | 482   | ns  | 0.33 |
| (2R,5R)-2-Methyl-5-(prop-1-en-2-yl)-2-vinyltetrahydrofuran | 1211| 1237 | 778.3 | 1415 | 925  | 814  | 1660 | 1204       | 932   | ns  | 0.23 |
| α-Mycene                          | 1162| 1164   | 894.4      | 956       | 973  | 1158 | 1378 | 1116       | 497   | ns  | 0.21 |
| 2-(1-Hexyn-1-yl)-3-(methoxymethyl)oxirane | 1325| na  | 735.3      | 504       | 962  | 1802 | 597  | 967        | 622   | ns  | 0.19 |
Table 2. Cont.

| Compound      | RI  | RI-Ref | Similarity | Volatile Composition (Area Counts) | F Prob  | %  |
|---------------|-----|--------|------------|------------------------------------|---------|----|
|               |     |        |            | NS | PE | NB | QC | Grand Mean | SEM |     |     |
| Limonene      | 1202| 1198   | 908.8      | 543| 580| 983| 1394| 875 | 359 | ns  | 0.17|
| Eucalyptol    | 1209| 1225   | 880        | –  | 421| 2375| –  | 699 | 298 | 0.004| 0.13|
| β-Ocimene     | 1252| 1248   | 898        | 1077|428| 702| 582| 697 | 325 | ns  | 0.13|
| Terpinolene   | 1286| 1281   | 906.6      | 651| 402| 851| 788| 673 | 377 | ns  | 0.13|
| p-Cymenene    | 1444| 1439   | 933.1      | 754| 478| 774| 648| 663 | 276 | ns  | 0.13|
| Total         |     |        |            | 20,680| 16,742| 28,414| 26,617| 23,113| 9409 | ns  | 4.43|
| Grand Total   |     |        |            | 615,776| 396,284| 541,546| 531,525| 521,283| 255,543| ns  | 100.0|

1 Values represent the mean of 3 commercial fields for each cultivar (n = 3). Only compounds with an average abundance >0.05% are shown. 2 Reference RI values are the average of 3 or more values from the National Institute of Standards and Technology (NIST) 2017 RI Database unless indicated otherwise. 3 MS Similarity values are the average of 3 samples unless indicated otherwise. 4 Significance effects among provinces based on ANOVA. 5 Value was below the threshold relative abundance of 0.05%. 6 Based on 1 value. 7 Based on 2 values. 8 na-RI not available in published databases.
The C<sub>6</sub> alcohols and aldehydes were abundant in wild lowbush blueberry fruit and contributed strong aromas (Tables 1 and 2). (Z)-3-Hexen-1-ol contributed a “green-grassy” aroma as did hexanal and (Z)-3-hexenal. (E)-2-Hexen-1-ol was described as having a “rancid-cheesy” aroma, while (E)-2-hexenal was described as “floral” and “sweet”. These three C<sub>6</sub> aldehydes comprised over 21% of the total volatiles in wild lowbush fruit, while the C<sub>6</sub> alcohols accounted for <2%, but were greater contributors to the fruit aroma. (E)-2-Hexanal and (Z)-3-hexanol were previously reported in juice extracted from lowbush blueberry fruit [27]. However, none of these alcohols or aldehydes were found in headspace collected from whole lowbush blueberry fruit [26], suggesting that their formation occurred as a result of fruit homogenization. These C<sub>6</sub> alcohols and aldehydes are known to be products of lipoxygenase (LOX) activity, which occurs as a result of homogenization and the addition of NaCl to blueberry homogenates reduces LOX activity [28,29]. However, differences between headspace volatile profiles of whole fruit and fruit homogenized with NaCl suggests that inhibition of LOX activity by NaCl is not absolute. Other alcohols that contributed to the aroma of wild lowbush fruit included 2-ethyl-1-hexanol that contributed “sweet” and “floral” notes and was the most abundant alcohol comprising 5.2% of the total volatiles. 1-Pentanol and (E)-2-nonenal also contributed to the aroma of wild lowbush blueberry fruit (Tables 1 and 2).

Monoterpenoids that contributed to the aroma of wild lowbush blueberries were linalool and geraniol. They had similar average MF values of 45.5% and 43.4%, respectively, and both contributed “floral” and “sweet” aromas (Table 1). While both had similar contribution to aroma, linalool was found in much higher concentrations comprising over 2% of the total volatiles, while geraniol comprised <0.05%, suggesting it may have a lower odor threshold than linalool (Table 2). Cometto-Muñiz et al. [30] reported a lower odor threshold for geraniol (0.1 ppm) compared to linalool (1.0 ppm); however, other studies have not confirmed this difference [31].

In addition to 3-hexanone, several other ketones also contributed to wild lowbush blueberry fruit aroma that included 1-octen-3-one, 2-dodecanone, 1-penten-3-one and 3-heptanone (Table 1). 1-Octen-3-one coeluted with the ester methyl 2-pentenoate and had an MF value of 49%. This aroma-active peak was described as “earthy-musty” with fewer descriptors of “fruity”, which may reflect the contribution of methyl 2-pentenoate in this coelution. 2-Dodecanone had similar aroma strength and contributed “floral”, “fruity”, “citrus” and “herb-like” notes. 1-Penten-3-one and 3-heptanone were both described as “earthy-musty” and had less contribution to fruit aroma. None of these ketones have been previously reported in wild lowbush blueberry fruit.

3.2. Aroma-Active Volatiles of Cultivated Highbush Blueberries

Among the five highbush cultivars evaluated in this study, a total of forty-two peaks were identified through GC-O analysis with MF values > 25% in at least one cultivar (Table 3). ‘Duke’, ‘Brigitta’, ‘Jersey’, ‘Liberty’ and ‘Aurora’ each had twenty-six, twenty-one, fourteen, twenty-two and twenty-eight aroma-active peaks, respectively. Eighteen of the identified compounds comprising these peaks were determined to be monoterpenoids, nine aldehydes, seven ketones, five alcohols, four branched-chain esters, one straight-chain ester, two acids, one hydrocarbon and one unknown. Differences in aroma-active compounds among the five highbush cultivars were substantial. Of the forty-two peaks identified through GC-O, only seven had MF values > 25% in all five cultivars and twelve peaks were detected by GC-O in only a single cultivar (Table 3). Similar differences in aroma-active compounds among four Southern highbush blueberry cultivars were reported, where twenty-four of forty-three aroma-active compounds were found in all four cultivars [10].
Table 3. Aroma-active compounds in cultivated highbush blueberry fruit of the cultivars ‘Duke’, ‘Brigitta’, ‘Jersey’, ‘Liberty’ and ‘Aurora’ identified through gas chromatography-olfactometry (GC-O) and ranked by average modified frequency (MF) values.

| Compound                        | RI   | Duke | Brigitta | Jersey | Liberty | Aurora | Ave  | Aroma Descriptors                                                                 | ID Basis              |
|---------------------------------|------|------|----------|--------|---------|--------|------|-----------------------------------------------------------------------------------|-----------------------|
| Geraniol                        | 1827 | 68.0 | 69.7     | 75.7   | 74.0    | 66.1   | 70.7 | Floral (38), Citrus (18), Fruity (18), Sweet (15), Blueberry (11)                 | RI, Std, AD, MS       |
| (Z)-3-Hexen-1-ol                | 1378 | 70.6 | 65.3     | 61.0   | 64.5    | 72.1   | 66.7 | Green-grassy (20), Earthy-musty (18), Herb-like (16), Rancid-cheesy (14), Floral (13) | RI, Std, AD, MS       |
| 1-Octen-3-one                   | 1303 | 60.2 | 58.5     | 66.3   | 61.1    | 64.5   | 62.1 | Earthy-musty (19), Mushroom (13), Pungent-sharp (11), Herb-like (10), Rancid-cheesy (9) | RI, Std, AD, MS       |
| α-Terpineol                     | 1720 | 51.4 | 64.8     | 46.2   | 58.4    | 65.7   | 57.3 | Floral (20), Herb-like (16), Green-grassy (14), Citrus (11), Fruity (9)           | RI, Std, AD, MS       |
| Linalool                        | 1531 | 65.3 | 46.5     | 66.9   | 44.7    | 55.1   | 55.7 | Floral (19), Fruity (18), Sweet (18), Herb-like (9), Citrus (8)                    | RI, Std, AD, MS       |
| 2-Ethyl-1-hexanol               | 1495 | 46.5 | 59.3     | 69.7   | 52.5    | 31.5   | 51.9 | Floral (18), Sweet (11), Fruity (8), Citrus (8), Green-grassy (7), Herb-like (7) | RI, Std, AD, MS       |
| 2-Undecanone                    | 1592 | 37.7 | 44.2     | 33.7   | 55.9    | 44.7   | 43.3 | Earthy-musty (5), Rancid-cheesy (4), Pungent-sharp (4), Blueberry (4), Roasted-nutty (4) | RI, Std, AD, MS       |
| (E)-2-Hexenal/α-Ocimene         | 1228 | 34.8 | 45.6     | 62.0   | 72.1    | 42.9   | 42.9 | Sweet (16), Floral (14), Fruity (12), Citrus (5), Green-grassy (5)               | RI, Std, AD, MS/      |
| Ethyl 3-methylbutanoate         | 1067 | 37.0 | 50.4     | 26.8   | 60.2    | 42.4   | 42.4 | Sweet (15), Blueberry (14), Fruity (14), Floral (6)                               | RI, Std, AD, MS       |
| Hexanal                         | 1088 | 40.0 | 60.8     | 49.3   | 62.0    | 42.4   | 42.4 | Green-grassy (28), Floral (8), Herb-like (6), Fruity (4), Sweet (2)              | RI, Std, AD, MS       |
| (E)-2-Hexen-1-ol/2,6-Dimethyl-2,6-octadiene | 1401 | 58.2 | 54.8     | 42.2   | 34.8    | 38.0   | 38.0 | Rancid-cheesy (16), Roasted-nutty (9), Earthy-musty (7), Sulfury (5), Blueberry (3), Herb-like (3) | RI, Std, AD, MS/      |
| Ethyl 2-methylbutanoate         | 1051 | 33.5 | 30.6     | 61.1   | 62.7    | 37.6   | 37.6 | Sweet (16), Fruity (15), Blueberry (8), Floral (7), Citrus (2), Herb-like (2)     | RI, Std, AD, MS       |
| 1-Pentanol                      | 1244 | 40.4 | 52.5     | 46.2   | 45.4    | 36.9   | 36.9 | Herb-like (7), Rancid-cheesy (7), Earthy-musty (5), Citrus (5), Pungent-sharp (4), Roasted-nutty (4), Sweet (4) | RI, Std, AD, MS       |
| Nonanal                         | 1395 | 33.5 | 59.3     | 43.2   | 45.6    | 36.3   | 36.3 | Herb-like (5), Fruity (2), Sweet (2), Floral (2), Citrus (2), Pungent-sharp (2)   | RI, Std, AD, MS       |
| Methyl 2-methylbutanoate        | 1014 | 24.2 | 48.1     | 69.7   | 37.7    | 35.9   | 35.9 | Fruity (22), Sweet (17), Blueberry (8), Floral (6), Citrus (3), Carmel (3)        | RI, Std, AD, MS       |
Table 3. Cont.

| Compound                                                                 | RI  | Duke | Brigitta | Jersey | Liberty | Aurora | Ave  | Aroma Descriptors 2                                      | ID Basis 3       |
|--------------------------------------------------------------------------|-----|------|----------|--------|---------|--------|------|--------------------------------------------------------|------------------|
| (Z)-3-Hexenal                                                           | 1151| 39.0 | 48.3     | 42.1   | 48.1    | 35.5   |      | Green-grassy (22), Floral (9), Herb-like (7), Fruity (6), Citrus (4) | RI, Std, AD, MS   |
| 2-Nonanone                                                               | 1384| 56.6 | 17.9     | 48.2   | 39.8    | 32.5   |      | Sweet (13), Fruity (12), Floral (10), Rancid-cheesy (6), Blueberry (5), Earthy-musty (5) | RI, Std, AD, MS   |
| (2R,5S)-2-Methyl-5-(prop-1-en-2-yl)-2-vinyltetrahydrofuran 5/β-Phellandrene 5/Eucalyptol | 1212| 42.9 | 42.9     | 52.3   | 24.2    | 32.5   |      | Floral (11), Sweet (7), Fruity (6), Citrus (6), Herb-like (4), Green-grassy (4), Rancid-cheesy (4) | RI, Std, AD, MS/ | RI, Std, AD, MS/ | RI, Std, AD, MS/ | RI, Std, AD, MS/ | RI, Std, AD, MS/ | RI, Std, AD, MS/ | RI, Std, AD, MS/ | RI, Std, AD, MS/ | RI, Std, AD, MS/ |
| 1-Penten-3-one/Ethyl butanoate                                           | 1031| 21.9 | 24.0     | 39.8   | 67.9    | 30.7   |      | Roasted-nutty (7), Herb-like (5), Earthy-musty (5), Fruity (4), Floral (3), Green-grassy (3), Rancid-cheesy (3) | RI, Std, AD, MS   |
| (E)-2-Nonenal                                                           | 1540| 29.2 | 44.7     | 52.5   | 25.3    |        |      | Earthy-musty (7), Rancid-cheesy (6), Roasted-nutty (5), Forest soil (4), Blueberry (3), Floral (3), Chemical (3), Herb-like (3) | RI, Std, AD, MS   |
| 2,6,6-Trimethyl-2-vinyltetrahydropyran 5                                 | 1111| 29.2 | 40.0     | 35.8   | 18.9    | 24.8   |      | Fruity (5), Earthy-musty (5), Herb-like (4), Rancid-cheesy (3), Roasted-nutty (3), Green-grassy (3), Citrus (3) | RI, Std, AD, MS   |
| 2-Heptanone                                                             | 1180| 31.0 | 33.5     | 49.4   | 22.8    |        |      | Sweet (6), Floral (6), Green-grassy (5), Fruity (3), Earthy-musty (3), Roasted-nutty (3) | RI, Std, AD, MS   |
| (E,E)-2,4-Heptadienal 5/(E)-Linalool oxide                              | 1468| 24.5 | 53.7     | 29.5   | 21.5    |        |      | Roasted-nutty (9), Rancid-cheesy (8), Earthy-musty (2), Blueberry (2), Sweet (6), Fruity (5), Rancid-cheesy (4), Pungent-sharp (3), Blueberry (2), Caramel (2), Earthy-musty (2) | RI, Std, AD, MS   |
| Methyl 3-methylbutanoate                                                | 1021| 43.8 | 56.6     |        |        |        |      | Rancid-cheesy (9), Earthy-musty (7), Pungent-sharp (5), Sweet (2) | RI, Std, AD, MS   |
| Unknown 787                                                             | 787 | 55.9 | 34.4     |        |        |        |      | Rancid-cheesy (9), Earthy-musty (7), Pungent-sharp (5), Sweet (2) | RI, Std, AD, MS   |
| (E)-2-Heptenal                                                          | 1330| 33.5 | 50.4     |        |        |        |      | Floral (4), Fruity (4), Citrus (4), Sweet (3), Roasted-nutty (3) | RI, Std, AD, MS   |
| Decanal                                                                 | 1491| 24.5 | 24.5     | 31.6   |        |        |      | Earthy-musty (5), Rancid-cheesy (3), Herb-like (3), Fruity (2), Floral (2), Green-grassy (2), Sweet (2) | RI, Std, AD, MS   |
| 2,2-Dimethyl propanoic acid 5                                           | 1572| 25.3 | 24.5     |        |        | 10.0   |      | Citrus (1), Blueberry (1), Carmel (1), Pungent-sharp (1), Roasted-nutty (1), Animal (1), cooked chicken (1), Acidic-vinegar (1), Earthy-musty (1), Herb-like (1) | RI, Std, MS       |
Table 3. Cont.

| Compound                        | RI   | MF Value (%) | Aroma Descriptors 2 | ID Basis 3 |
|---------------------------------|------|--------------|---------------------|------------|
|                                 |      | Duke | Brigitta | Jersey | Liberty | Aurora | Ave |                   |             |
| D-Carvone                       | 1752 | 25.3 | 24.2    | 9.9    |          |        |     | Floral (4), Fruity (3), Roasted-nutty (3), Herb-like (2), Green-grassy (2), Sweet (1) Acidic-vinegar (1), Earthy-musty (1), Rancid-cheesy (1) | RI, Std, AD, MS |
| Acetophenone                    | 1643 | 45.6 | 9.1     |        |          |        |     | Earthy-musty (2), Roasted-nutty (1), Chemical (1), Burnt Toast (1), Burning grass (1), Rancid-cheesy (1) Floral (4), Roasted-nutty (3), Herb-like (3), | RI, Std, AD, MS |
| 2-Dodecanone                    | 1711 | 44.7 | 8.9     |        |          |        |     | Green-grassy (1), Pungent-sharp (1), Rancid-cheesy (1) Roasted-nutty (2), Fruity (2), Sweet (1), Floral (1), | RI, Std, AD, MS |
| 2-Methyl-1,4-pentadiene 5       | 1100 | 36.9 | 7.4     |        |          |        |     | Herb-like (1), Pungent-sharp (1), Ground coffee (1), Green-grassy (1), Chemical (1) | RI, MS |
| (E)-Isopiperitenol 5            | 1769 | 29.5 | 7.3     | 7.4    |          |        |     | Sweet (3), Blueberry (2), Citrus (1), Fruity (1), Floral (1), Blackberry (1), Herb-like (1), Roasted-nutty (1) | RI, AD, MS |
| Geranyl acetone 5               | 1863 | 34.8 | 7.0     |        |          |        |     | Fruity (3), Sweet (2), Roasted-nutty (2), Floral (1), Citrus (1), Blueberry (1), Earthy-musty (1), Horse (1) | RI, AD, MS |
| 2,3,6-Trimethyl-1,5-heptadiene 5| 1407 | 34.4 | 6.9     |        |          |        |     | Floral (4), Sweet (2), Fruity (2), Earthy-musty (2), | RI, MS |
| α-Phellandrene 1                | 1170 | 32.7 | 6.5     |        |          |        |     | | RI, Std, AD, MS |
| Nerol                           | 1797 | 29.5 |        | 5.9    |          |        |     | Sweet (3), Floral (3), Citrus (2), Herb-like (2), Dill (1), Roasted, nutty (3), Floral (2), Rose (1), Sweet (1), Fruity (1), | RI, Std, AD, MS |
| Anethofuran 5                   | 1509 | 29.2 | 5.8     |        |          |        |     | Earthy, musty (1), Camp Fire (1), Herb-like (1), Rancid, cheesy (1) | RI, AD, MS |
| (E)-3-Hexen-1-ol                | 1355 | 28.0 | 5.6     |        |          |        |     | Sweet (2), Citrus (2), Fruity (2), Rancid-cheesy (2), Rancid, cheesy (2), Fruity (1), Blueberry (1) | RI, Std, AD, MS |
| Octanal                         | 1290 | 26.7 | 5.3     |        |          |        |     | Rancid-cheesy (3), Citrus (2), Roasted-nutty (1), Sweet(1), Floral (1), Earthy-musty (1) | RI, Std, AD, MS |
| Limonene                        | 1203 | 25.8 | 5.2     |        |          |        |     | Earthy-musty (2), Sweet (2), Green-grassy (1), Floral (1), Fruity (1), Rancid-cheesy (1), Earthy-musty (1), Sulfur (1), Chemical cleaner (1) | RI, Std, AD, MS |
| 5-Methylhexanoic acid           | 1906 | 25.3 |        | 5.1    |          |        |     | | RI, Std, AD, MS |

1 Values for each cultivar are the average of fifteen GC-O analyses conducted on fruit from three commercial fields by five evaluators for each sample. Compounds presented had MF values > 25% in at least one cultivar. 2 The five most frequent descriptors. 3 RI, retention index; Std, standard; AD, aroma description; MS, mass spectrum (See Table 4 for RI reference comparisons and MS similarity values). 4 Frequency of descriptor chosen by GC-O panelists. 5 Tentative identification.
Table 4. Headspace volatile composition of cultivated highbush blueberry fruit of the cultivars ‘Duke’, ‘Brigitta’, ‘Jersey’, ‘Liberty’ and ‘Aurora’ determined by 2-dimensional gas chromatography–time of flight–mass spectrometry (GCxGC-TOF-MS) 1.

| Compound          | RI   | RI-Ref | Sim  | Volatile Composition (Area Counts) | F Prob | %   |
|------------------|------|--------|------|-----------------------------------|--------|-----|
|                   |      |        |      | Duke | Brigitta | Jersey | Liberty | Aurora | Mean | SEM |
| Acids             |      |        |      |      |          |        |         |        |      |     |
| 2-Ethylhexenoic acid | 1935 | 1952   | 933  | 5    | 227      | 306    | –        | 442    | 195  | 191 |
| Total             |      |        |      |      | 227      | 306    | –        | 442    | 195  | 191 |
| Alcohols          |      |        |      |      |          |        |         |        |      |     |
| (E)-2-Hexen-1-ol  | 1404 | 1402   | 933  | 4509 | 2895     | 1493   | 4395     | 4530   | 3564 | 646 |
| (E)-2-Hexen-4-yn-1-ol | 1221 | na 6   | 856  | 1854 | 1604    | 685    | 2651     | 4001   | 2159 | 718 |
| 1-Hexanol         | 1347 | 1373   | 923  | 2141 | 1058    | 1531   | 2447     | 2324   | 1900 | 249 |
| 2-Ethyl-1-hexanol | 1487 | 1492   | 924  | 1885 | 1827    | 1468   | 538      | 3080   | 1760 | 1034|
| 1-Pentanol        | 1245 | 1242   | 896  | 636  | 1514    | 1298   | 1414     | 634    | 1099 | 221 |
| 2-Hexyn-1-ol      | 1205 | 1207   | 866  | 993  | 960     | 643    | 1418     | 1174   | 1038 | 349 |
| (Z)-3-Hexen-1-ol  | 1383 | 1381   | 941  | 1177 | 149     | 1195   | 835      | 540    | 779  | 143 |
| 2-Butanol         | 1016 | 1020   | 843  | 221  | 188     | 329    | 638      | 449    | 365  | 69  |
| 1-Octen-3-ol      | 1446 | 1443   | 868  | 141  | 288     | 215    | 242      | 266    | 230  | 53  |
| Cyclobutanol      | 1043 | NA     | 789  | 131  | 198     | 172    | 131      | 284    | 183  | 51  |
| Total             |      |        |      | 13,689 | 10,680 | 9028   | 14,709   | 17,282 | 13,185 | 6.69 |
| Aldehydes         |      |        |      |      |          |        |         |        |      |     |
| (E)-2-Hexenal     | 1222 | 1216   | 941  | 49,150 | 49,217 | 22,884 | 69,553   | 73,969 | 52,955 | 6302 |
| Hexanal           | 1083 | 1087   | 905  | 29,102 | 14,807 | 14,784 | 21,079   | 18,948 | 19,744 | 2270 |
| (Z)-3-Hexenal     | 1148 | 1146 7 | 887  | 31,965 | 1581   | 8590   | 9550     | 7589   | 11855 | 3119 |
| Pentanal          | 979  | 981    | 899  | 1800 | 7104    | 1710   | 5148     | 4975   | 4148  | 1065|
| (E,E)-2,4-Hexadienal | 1405 | 1401   | 926  | 2022 | 469     | 636    | 1162     | 1068   | 1071  | 201 |
| (E)-3-Hexenal     | 1137 | 1138   | 866  | 1302 | 745     | 732    | 886      | 878    | 909   | 232 |
| Heptanal          | 1186 | 1184   | 909  | 274  | 1161    | 425    | 697      | 1338   | 779   | 231 |
| Nonanal           | 1396 | 1389   | 897  | 990  | 621     | 298    | 331      | 233    | 494   | 276 |
| Methacrolein      | 881  | 890    | 844  | 113  | 610     | 187    | 333      | 589    | 366   | 110 |
| 4-Pentenal        | 1133 | 1129   | 852  | 88   | 345     | 143    | 287      | 755    | 323   | 59  |
| (E)-2-Heptenal    | 1329 | 1323   | 922  | 319  | 388     | 202    | 212      | 318    | 288   | 109 |
| Octanal           | 1291 | 1284   | 919  | 249  | 196     | 75     | 244      | 153    | 90    | 80  |
| (E)-2-Octenal     | 918  | 913    | 835  | 30   | 176     | 82     | 263      | 110    | 64    | 64  |
| 3-Methylbutanal   | 1435 | 1436   | 909  | 81   | 81      | 130    | 35       | 116    | 89    | 53  |
| Total             |      |        |      | 117,485 | 77,501 | 50,721 | 109,430  | 111,282 | 95,184 | 9472 |

1. Data presented as mean ± SEM. ns indicates not significant.
Table 4. Cont.

| Compound                      | RI     | RI-Ref | Sim | Volatile Composition (Area Counts) | F Prob | %  |
|-------------------------------|--------|--------|-----|------------------------------------|--------|----|
|                               |        |        |     | Duke | Brigita | Jersey | Liberty | Aurora | Mean | SEM |        |        |
| Esters-Branched Chain         |        |        |     |      |         |        |         |        |      |     |        |        |
| Methyl 3-methylbutanoate      | 1018   | 1016   | 933 | 1803 | 5783 | 12664 | –       | 1332  | 4316 | 826 | <0.001 | 2.21  |
| Methyl 2-methylbutanoate      | 1010   | 1008   | 896 | –    | 563  | 1012  | –       | –    | 315  | 68  | <0.001 | 0.16  |
| Ethyl 3-methylbutanoate       | 1065   | 1066   | 942 | 7    | 177  | 398   | –       | –    | 116  | 48  | <0.001 | 0.06  |
| Total                         | 1809   | 6523   | 14,073 | –   | 1332 | 4895  | 1062    | –    |     |    | <0.001 | 2.43  |
| Esters-Straight Chain         |        |        |     |      |         |        |         |        |      |     |        |        |
| Ethyl acetate                 | 894    | 893    | 866 | 331  | 74   | 3093  | –       | –    | 699  | 133 | <0.001 | 0.36  |
| Methyl acetate                | 858    | 856    | 857 | –    | 1639 | 1288  | –       | 409  | 667  | 140 | <0.001 | 0.34  |
| (Z)-2-Hexen-1-ol acetate     | 1334   | 1329   | 881 | 1397 | 337  | 80    | 665     | 539  | 604  | 116 | <0.001 | 0.31  |
| Total                         | 1727   | 2051   | 4461 | 165  | 665  | 948   | 1887    | 320  |     |    | <0.001 | 1.01  |
| Furans                        |        |        |     |      |         |        |         |        |      |     |        |        |
| 2-Ethylfuran                  | 951    | 950    | 918 | 364  | 89   | 220   | 393     | 481  | 309  | 93  | 0.06   | 0.16  |
| Tetrahydrofuran               | 880    | 862    | 850 | 46   | –    | 242   | 422     | 185  | 179  | 20  | <0.001 | 0.09  |
| Total                         | 410    | 89     | 462 | 816  | 666  | 535   | 122     | 0.01 |     |    | 0.01  | 0.25  |
| Hydrocarbons                  |        |        |     |      |         |        |         |        |      |     |        |        |
| Hexane                        | 594    | 600    | 852 | 10127 | 7083 | 8141  | 10429   | 8238 | 8804 | 4146 | ns   | 4.50  |
| Ethylcyclobutane              | 809    | 692    | 886 | 2079 | 1936 | 1980  | 1994    | 1986 | 1995 | 712  | ns   | 1.02  |
| (Z,Z)-2,4-Hexadiene          | 738    | na     | 933 | 366  | 51   | 45    | 260     | 383  | 285  | 107 | 0.003 | 0.15  |
| Toluene                       | 1048   | 1053   | 916 | 133  | 121  | 605   | –       | –    | 172  | 40  | <0.001 | 0.09  |
| (E,Z)-2,4-Hexadiene          | 754    | na     | 937 | 23   | 205  | 62    | 286     | 157  | 147  | 73  | ns   | 0.08  |
| Total                         | 13,048 | 9395   | 10,834 | 12,969 | 10,765 | 11,463 | 4888    | ns   |     |    | 5.83  |        |
| Ketones                       |        |        |     |      |         |        |         |        |      |     |        |        |
| 2-Butanone                    | 905    | 903    | 916 | 21,885 | 5006 | 16,128 | 14,576  | 8948 | 13,309 | 854 | <0.001 | 6.81  |
| 6-Methyl-5-hepten-2-one       | 1338   | 1340   | 884 | 3923 | 2772 | 657   | 2336    | 1820 | 2302 | 458 | 0.002 | 1.18  |
| 3-Ethylidene-1-methoxy-5-hexen-2-one | 1325 | na | 731 | 5929 | 119 | 43 | 1568 | – | 1532 | 51 | <0.001 | 0.78  |
| Acetone                       | 852    | 836    | 953 | 354  | 406  | 377   | 554     | 348  | 408  | 48  | 0.043 | 0.21  |
| 1-Penten-3-one                | 1022   | 1032   | 878 | 164  | 484  | 134   | 332     | 506  | 324  | 98  | 0.04  | 0.17  |
| 2-Heptanone                   | 1182   | 1180   | 927 | 843  | 44   | 26    | 346     | 195  | 291  | 143 | 0.005 | 0.15  |
| 3-Methyl-3-buten-2-one        | 995    | 997    | 935 | 155  | 368  | 161   | 61      | 679  | 285  | 109 | 0.006 | 0.15  |
| 1-Octen-3-one                 | 1304   | 1301   | 842.5 | 35  | 255  | –     | 58      | 219  | 113  | 71  | 0.066 | 0.06  |
| Total                         | 33,287 | 9454   | 17,526 | 19,832 | 12,715 | 17,923 | 1963    | <0.001 |     |    | 9.50  |        |
| Compound                                                                 | RI   | RI-Ref² | Sim³ | Volatile Composition (Area Counts) |  | F Prob ⁴ | %    |
|-------------------------------------------------------------------------|------|---------|------|------------------------------------|---|----------|------|
| **Monoterpenoids**                                                      |      |         |      |                                    |   |          |      |
| Linalool                                                                | 1542 | 1540    | 902  | 48,358 8419 7395 15,063 9012 17,649 1408 | <0.001 | 9.03     |
| Linalool acetate                                                        | 1543 | 1548    | 866  | 31,870 – 34 30 6387 23             | <0.001 | 3.27     |
| α-Terpineol                                                             | 1698 | 1690    | 933  | 11,103 1428 2097 3456 2993 4215 1696 | 0.005   | 2.16     |
| (2R,5R)-2-Methyl-5-(prop-1-en-2-yl)-2-vinyltetrahydrofuran             | 1243 | 1233    | 905  | 12,803 216 105 2565 96 3157 94     | <0.001 | 1.61     |
| Limonene                                                                | 1211 | 1226    | 892  | 11,375 437 543 2500 917 3154 1118   | <0.001 | 1.61     |
| β-Myrcene                                                               | 1201 | 1226    | 885  | 891 533 540 1228 707 2280 1119     | <0.001 | 1.17     |
| (2R,5S)-2-Methyl-5-(prop-1-en-2-yl)-2-vinyltetrahydrofuran             | 1162 | 1164    | 886  | 9476 – 624 – 2020 75              | <0.001 | 1.03     |
| α-Ocimene                                                               | 1234 | 1238    | 900  | 6264 334 233 798 383 1602 889     | <0.001 | 0.82     |
| Terpinolene                                                             | 1210 | 1204    | 850  | 6576 18 46 644 158 1489 1024     | <0.001 | 0.76     |
| Eucalyptol                                                              | 1285 | 1280    | 898  | – 4650 250 2417 38 1471 245     | <0.001 | 0.75     |
| 1,3,8-p-Menthatriene                                                    | 1234 | 1242    | 912  | 5427 – 1175 – 1320 239            | <0.001 | 0.68     |
| Dehydro-p-cymene                                                       | 1616 | 1627⁷   | 888  | 4424 – 42 456 – 984 67           | <0.001 | 0.50     |
| β-Ocimene                                                               | 1443 | 1440    | 915  | 3370 175 121 468 279 883 481     | <0.001 | 0.45     |
| α-Cymene                                                               | 1276 | 1284    | 936  | 2316 – 258 0 515 5              | <0.001 | 0.26     |
| Geraniol                                                               | 1844 | 1851    | 855  | 1614 44 78 314 336 477 247      | 0.001  | 0.24     |
| α-Terpinene                                                            | 1182 | 1178    | 858  | 1988 – 193 0 436 306            | <0.001 | 0.22     |
| 2,6,6-Trimethyl-2-vinyltetrahydropyran                                  | 1111 | 1112    | 827  | 906 – 57 1094 411 189           | <0.001 | 0.21     |
| (E)-Geranylacetone                                                     | 1849 | 1837    | 820  | 364 900 – 549 191 401 77        | <0.001 | 0.21     |
| (E)-Dihydrocarvone                                                     | 1616 | 1627⁷   | 888  | 1594 – – 319 0              | ns    | 0.16     |
| Unknown 1611                                                           | 1611 | na      | 797  | 65 494 – 270 396 245 119      | 0.041  | 0.13     |
| 2,6-Dimethyl-2,6-octadiene                                              | 1405 | na      | 797  | 65 494 – 270 396 245 119      | 0.041  | 0.13     |
| γ-Terpinene                                                            | 1248 | 1241    | 864  | 1125 – 25 – 230 191            | 0.002  | 0.12     |
| α-Phellandrene                                                         | 1167 | 1172    | 890  | 1125 – – 225 0             | ns    | 0.12     |
| P-Cymene-8-ol                                                          | 1845 | 1839    | 859  | 705 – – 141 0              | ns    | 0.07     |
| Z-Linalool oxide                                                       | 1519 | 1513    | 873  | 91 – 442 – 107 13            | <0.001 | 0.05     |
| β-Phellandrene                                                         | 1212 | 1212    | 869  | 39 – 130 25 39 19           | <0.001 | 0.02     |
| **Total**                                                               |      |         |      |                                    | 172,402 | 17,647  | 11,448 | 34,047 | 16,655 | 48,273 | 11,115 | <0.001 | 25.80 |
Table 4. Cont.

| Compound                   | RI    | RI-Ref 2 | Sim 3 | Volatile Composition (Area Counts) | F Prob 4 | %   |
|----------------------------|-------|----------|-------|------------------------------------|----------|-----|
|                            |       |          |       | Duke | Brigita | Jersey | Liberty | Aurora | Mean | SEM |       |       |     |
| Total                      | 6337  |          |       | 752  | 49     | <0.001 | 0.65    |
| Dimethyl trisulfide        | 1391  | 1390     | 903   | 3759 | –       | –       | –       | –      | 752  | 49  | <0.001 | 0.38 |
| Dimethyl disulfide         | 1079  | 1069     | 944   | 2578 | –       | –       | –       | –      | 516  | 394 | <0.001 | 0.26 |
| Grand Total                | 360,259 | 133,672 | 118,910 | 192,484 | 172,128 | 206,647 | 26,301 | <0.001 | 100 |

1 Values are means of 2 harvests from 3 commercial fields for each cultivar (n = 6). Only compounds with an average relative abundance >0.05% are shown. 2 Reference RI values are the average of 3 or more values from the NIST 2017 RI Database unless indicated otherwise. 3 MS Similarity values are the average of 3 samples unless indicated otherwise. 4 Significance of effects among cultivars based on ANOVA. 5 Value was below the threshold relative abundance of 0.01%. 6 na-RI not available in published databases. 7 Based on 1 value. 8 Based on value.
Monoterpenoids were major contributors of "floral", "sweet", "fruity", "citrus" and "blueberry" aromas in fruit of the five highbush blueberry cultivars assessed in this study (Table 3). Of all the volatile compounds identified in these highbush fruits, monoterpenoids comprised, on average, over 25% of the total volatile content (Table 4). Geraniol, α-terpineol and linalool contributed strong aromas to the fruit of all five cultivars (Table 3). Linalool was the most abundant monoterpenoid comprising, on average, approximately 9% of the total volatiles, while α-terpineol comprised approximately 2% of the total volatiles. Geraniol averaged only 0.24% of the total volatiles but was the strongest contributor to aroma, having the first or second highest MF value for all five cultivars. Linalool, α-terpineol and geraniol were previously reported in highbush blueberries [12,17,20,29,32,33].

In four cultivars of Southern highbush fruit, linalool, and to a lesser extent, α-terpineol, contributed to the aroma of fruit, but geraniol only contributed to the aroma of two of these cultivars [10]. The occurrence of the remaining fifteen monoterpenoids that were identified through GC-O in this study varied among the five cultivars. With the exception of 2,6-dimethyl-2,6-octadiene, no additional monoterpenoids were identified in ‘Jersey’ fruit by GC-O. Six of the identified aroma-active monoterpenoids were only found in fruit of one of the five cultivars. Other studies reported differences in monoterpenoid composition among cultivars [14,19,33–36], and it was suggested that differences in monoterpenoid content could be responsible for the distinct aroma of different blueberry cultivars as well as consumer acceptability [14,36].

Branched-chain esters also contributed "fruity" and "sweet" aroma to highbush blueberry fruit, but their contribution varied among the five cultivars. Methyl 3-methylbutanoate, ethyl 2-methylbutanoate and methyl 2-methylbutanoate were strong contributors to the aroma of ‘Jersey’ fruit and moderate contributors to ‘Brigitta’ fruit (Table 3). In ‘Aurora’ fruit, ethyl 2-methylbutanoate and ethyl 3-methylbutanoate were strong contributors to the aroma. In contrast, the aroma from these esters was not detected in ‘Liberty’ fruit. The GC-MS volatile profiles of these fruit found methyl 3-methylbutanoate to be the most abundant branched-chain ester, followed by methyl 2-methylbutanoate, and they comprised approximately 12% and 5% of the total volatiles in ‘Jersey’ and ‘Brigitta’ fruit, respectively, but <1% in the other three cultivars (Table 4). Other studies have shown that the contribution of branched-chain esters to highbush blueberry flavor is cultivar dependent. Qian et al. [33] found high levels of branched-chain esters in ‘Duke’ fruit but low concentrations in ‘Aurora’ and ‘Liberty’. In both Northern and Southern highbush fruit, GC-O analysis identified that all four methylbutanoates contributed “fruity” aroma notes, although the contribution of each varied among cultivars [10,20]. In fruit from six highbush blueberry cultivars, GC-MS analysis detected methyl 2-methylbutanoate and ethyl 3-methylbutanoate in all six cultivars and ethyl 2-methylbutanoate in five of the six cultivars in low or trace concentrations, but methyl 3-methylbutanoate was not detected [29]. Qian et al. [33] suggested that branched-chain esters were associated with highbush cultivars that had desirable flavor.

The C6 alcohols and aldehydes were strong contributors to the aroma of cultivated highbush blueberry and comprised nearly half of the volatile compounds (Tables 3 and 4). (E)-2-Hexenal and hexanal comprised 27.1% and 10.1% of the total volatiles, respectively. However, (Z)-3-hexen-1-ol that comprised only 0.4% of total volatiles was the strongest contributor of aroma to fruit of all five cultivars based on MF values. (E)-2-Hexenal and hexanal, which were also strong aroma contributors, were not identified by GC-O in ‘Jersey’ fruit. Moreover, GC-O analysis did not detect (E)-2-hexen-1-ol in ‘Liberty’ fruit or (Z)-3-hexen in ‘Brigitta’ fruit. In two cultivars of Northern highbush fruit, GC-O identified all six of these C6 aldehydes and alcohols but only (Z)-3-hexen-1-al and (Z)-3-hexen-1-ol made a strong contribution to aroma [20]. In Southern highbush blueberries, three C6 aldehydes contributed to the aroma of all four cultivars, but of the C6 alcohols only (Z)-3-hexenol was detected by GC-O in one cultivar [10]. Horvat and Senter [37] reported that (E)-2-hexenal, (E)-2-hexenol and (Z)-3-hexenol were key components in blueberry flavor. Other alcohols and aldehydes that contributed strong or moderate aromas in fruit of
all five cultivars of highbush blueberries in this study included 2-ethyl-1-hexanol, which contributed “floral” and “sweet” aromas and 2,6-nonadienal, which contributed “green-grassy” and “floral” aromas. These compounds were previously identified in highbush blueberry fruit \[12,20,29\].

Ketones also contributed to the aroma of cultivated highbush blueberry fruit. The most aroma-active ketone present in all five cultivars was 1-octen-3-one, which contributed an “earthy-musty”, “mushroom-like” aroma (Table 3), but, on average, only accounted for 0.06% of total volatiles. 1-Octen-3-one was previously reported to contribute to the aroma of highbush fruit \[10,20\]. The ketones (E,Z)-2-undecanone, 2-nonanone, 2-heptanone and, to a lesser degree, 1-penten-3-one, contributed a variety of aromas including “floral” and “fruity” notes to the highbush blueberry fruit in this study; the former three compounds were previously reported in highbush blueberries \[10,12,20,29\].

3.3. Comparison of Aroma-Active Compounds in Wild Lowbush and Cultivated Highbush Blueberries

Many of the aroma-active compounds identified in wild lowbush blueberry fruit were also identified to contribute to the aroma of cultivated highbush fruit. Of the twenty-three aroma peaks found in wild lowbush blueberry fruit, nineteen were found in at least one of the five highbush cultivars assessed in this study. Aroma-active compounds that were unique to wild lowbush fruit in this study included the three esters ethyl propanoate, ethyl 2-methylpropanoate and methyl 3-methyl-2-butenoate; one alcohol 1-hexanol; and one ketone 3-heptanone. Conversely, of the forty-two aroma-active peaks identified in the five highbush cultivars, twenty-three were not found to contribute to the aroma of wild lowbush fruit. These peaks included sixteen monoterpenoids, five aldehydes, four ketones, two acids, one alcohol and one hydrocarbon. While there were many similarities in the aroma-active compounds in the fruit of these two species, the quantities of these compounds and their contribution to fruit aroma differed considerably. These differences would impact the flavor of blueberry products and should be considered in product formulation.

The volatile profile composition of wild lowbush blueberries was dominated by esters with branched-chain and straight-chain esters comprising 31.6% and 15.9% of the total volatiles, respectively (Figure 1, Table 2). The ester content of cultivated highbush blueberries was lower than that of wild lowbush fruit with branched-chain esters and straight-chain esters averaging 2.4% and 1.0% of total volatiles, respectively (Figure 1, Table 4). Aldehydes were the most abundant volatile group in highbush fruit comprising 47.7% of the total volatiles, while in wild lowbush fruit, they were the second most abundant group comprising 28.6% of the total volatiles. The second most abundant group of volatiles in highbush fruit was monoterpenoids comprising 25.8% of total volatiles. In lowbush fruit, monoterpenoids accounted for only 4.4% of the total volatiles. Similar concentrations of alcohols (8.8% vs. 6.7%), ketones (5.9% vs. 9.5%) and hydrocarbons (2.3% vs. 5.8%) were observed in the wild lowbush and cultivated highbush fruit, respectively.
Jersey' fruit, branched-chain esters (11.8%) and ketones (14.7%) were more abundant than
while 'Duke' differed and associated with monoterpenoid content.
volatile chemical groups (Figure 2). Scores 1 and 2 accounted for 75% of the variability.
volatile compounds among chemical groups were seen in cultivated highbush blueberry
procines in eastern Canada (n = 12) and five cultivars of cultivated highbush fruit collected from three
highbush blueberry fruit grouped according to chemical properties. Values represent the mean area
counts collected from wild lowbush fruit samples from three commercial fields located in four
provinces in eastern Canada (n = 12) and five cultivars of cultivated highbush fruit collected from
three fields and two harvests (n = 30).

Volatile composition among chemical groups in wild lowbush fruit did not differ
significantly among provinces except for straight chain esters (p = 0.016) that comprised
only 4.4% of total volatiles in fruit from PE compared to 13.6%, 23.6%, and 19.3% in fruit
from NS, NB and QC, respectively (Table 2). Greater differences in the distribution of
volatile compounds among chemical groups were seen in cultivated highbush blueberry
fruit (Table 4). Aldehyde composition varied significantly among cultivars (p ≤ 0.001),
comprising over half of 'Aurora', 'Brigitta' and 'Liberty' total volatiles, but <43% of total
volatiles in 'Duke' and 'Jersey' fruit. Monoterpenoid content ranged from 47.9% in 'Duke'
to 9.6% in 'Jersey' (p < 0.001). Pico et al. [29] also reported the fruit of 'Duke' to have
higher concentrations of total terpenes than the fruit of other highbush cultivars. In
'Jersey' fruit, branched-chain esters (11.8%) and ketones (14.7%) were more abundant than
monoterpenoids (9.6%). The average composition of other fruit volatiles that differed
significantly among the five cultivars studied included ketones, branched-chain esters,
straight-chain esters and furans. Qian et al. [33] found three selections from the USDA
blueberry breeding program that were considered to have “outstanding” flavor had higher
branch-chain ester content than seven highbush cultivars.

To further explore the differences in the volatile chemistry between wild lowbush
and cultivated highbush blueberries, principal component analysis was conducted on the
volatile chemical groups (Figure 2). Scores 1 and 2 accounted for 75% of the variability.
Score 1 was driven by alcohol, acid, straight-chain ester and total volatile content and score
2 was driven by branched-chain ester, hydrocarbon and ketone content. Differences were
seen between the wild lowbush and the cultivated highbush fruit, with the former found
in the right of the plot and the latter in the left. Wild lowbush fruit from NB and QC were
similar, while those from NS and PE differed reflecting differences in total volatiles, acids,
alcohols and esters. Among the highbush cultivars, 'Brigitta' and 'Jersey' were similar,
while 'Duke' differed and associated with monoterpenoid content.

Figure 1. The distribution of volatile compounds in the headspace of wild lowbush and cultivated
highbush blueberry fruit grouped according to chemical properties. Values represent the mean area
counts collected from wild lowbush fruit samples from three commercial fields located in four
provinces in eastern Canada (n = 12) and five cultivars of cultivated highbush fruit collected from
three fields and two harvests (n = 30).
These differences in volatile profiles were reflected in the differences in aroma-active compounds. Wild lowbush blueberry aroma was dominated by esters, while cultivated highbush fruit was dominated by monoterpenoids. In wild lowbush blueberry fruit, the five most aroma-active compounds were esters (Table 1), while in highbush fruit, three of the five were monoterpenoids and none were esters (Table 2). Lowbush fruit had two monoterpenoids that ranked eleventh and twelfth of the most aroma-active compounds, while esters in highbush fruit ranked ninth, twelfth, fifteenth and twenty-fourth, and each ester was not found in all cultivars. Alcohols, ketones and aldehydes all contributed similarly to the aroma of fruit from both blueberry species, although specific differences were observed in compounds and concentrations.

The method of volatile collection and analysis can affect volatile profiles and aroma-activity assessment [13,24,38]. All methods have advantages and disadvantages, and no method is ideal. In our study, headspace volatiles were collected from fruit homogenized in saturated salt using a DVB/CAR/PDMS SPME fiber, and aroma activity was accessed by GC-O using a sensory panel that consisted of nine evaluators. SPME fibers have a degree of selectivity and have good sensitivity for compounds with low molecular weight and high volatility but may underestimate those with low volatility [24,38]. In contrast, solvent extraction of fruit and concentration of volatiles using solvent extract dilution analysis (SAFE) is more effective in capturing volatile compounds with low volatility, including acids and hydroxyl-containing compounds. Using a total extract and SAFE, Qian et al. [20] reported vanillin as an odorant in highbush blueberry, but did not report the presence of vanillin when using SPME analysis [33]. However, the total extracts obtained by solvent extraction are less reflective of the head space volatile composition that induces the olfactory response of the consumer, and valid olfactory rankings are only obtained after odor activity values are calculated using odor thresholds for each compound [24]. In our study, aroma activity was assessed using a direct intensity measure that integrated an intensity rating and the detection frequency of panelists. The panel helps to account for variability in aroma sensitivity among individuals. Additional studies could be conducted using different analytical method such as SAFE and dilution analysis to further assess the contribution of volatiles to the aroma of wild lowbush blueberry.
To further compare the sensory impact of the aroma-active compounds of wild lowbush and cultivated highbush blueberry fruit, the frequency of descriptors chosen by sensory panelists to describe the aroma-active peaks was analyzed by ANOVA and illustrated using a radar plot (Figure 3). There was a significant interaction ($p < 0.001$) in the frequency of descriptors chosen between blueberry species and the descriptor. Of the 16 descriptors provided to the panelists, “floral” was chosen most frequently followed by “fruity” and “sweet” in both wild lowbush and cultivated highbush fruit. The fourth and fifth most frequent descriptors for wild lowbush fruit were “blueberry” and “green-grassy”, whereas for cultivated fruit, “green-grassy”, “herb-like”, “rancid-cheesy” and “earthy-musty” were chosen more frequently than “blueberry”. The descriptors “floral”, “fruity”, “sweet” and “blueberry” were chosen significantly more times to describe aroma-active compounds in wild lowbush than in cultivated highbush blueberry fruit. These results suggest that wild lowbush blueberries were perceived to have a fruitier and more “blueberry-like” aroma than cultivated highbush fruit in this study.

![Radar plot of descriptors chosen by sensory panelists to describe aroma-active compounds](image)

**Figure 3.** Frequency of descriptors chosen by sensory panelists to describe aroma-active compounds from wild lowbush and cultivated highbush blueberry fruit samples analyzed by gas chromatography-olfactometry (GC-O). Values represent the mean frequencies of descriptors chosen by five evaluators. Significant differences determined by LSD$_{0.05}$ are indicated by “*” next to the descriptor. GC-O analysis was conducted on fruit collected from three wild lowbush blueberry fields located in four provinces ($n = 12$), and from five cultivars of cultivated highbush blueberries from three fields ($n = 15$).

In addition to differences in volatile profiles that contribute to flavor differences between lowbush and highbush fruit, differences in sugar and acid composition may also impact flavor differences. Wild lowbush blueberry fruit have higher sugar content than cultivated highbush fruit, and the predominant acid is quinic acid compared to citric acid in highbush fruit [11]. Quinic acid has a less tart taste compared to citric acid [39], which would contribute to a sweeter less tart flavor of wild lowbush fruit compared to cultivated highbush fruit. The fruit of ‘Duke’, which has 4% *V. angustifolium* in its parentage, was reported to have higher quinic acid content than the other four cultivars in this study [11]. However, the volatile composition of ‘Duke’ did not show additional similarities to wild lowbush fruit.
4. Conclusions

The aroma-active compound composition of wild lowbush fruit produced among four Canadian provinces was more consistent than that found among the five highbush cultivars assessed in this study. Wild lowbush blueberry fields are made up of a complex mixture of genotypes that are naturally occurring clones. Genotypic variation in aroma volatile composition among wild clones can be expected. However, the genotypic diversity among the large number of wild clones that were commercially harvested for this study resulted in a fairly consistent aroma composition regardless of the province of production. Aroma-active volatiles in wild lowbush fruit were dominated by esters that contributed “fruity” and “sweet” aromas. This was in contrast to the variation in aroma volatile composition among highbush blueberry cultivars, which are each a unique genetic clone. Aroma-active volatiles in cultivated highbush fruit were dominated by monoterpenoids that contributed “floral” aromas. Frozen blueberry fruit marketed as ingredients for food products are typically marketed as “wild blueberries” or “blueberries” (cultivated highbush) with no identification of cultivar in the later. The greater homogeneity of volatile composition in wild lowbush fruit suggests that they would impart more consistent flavor characteristics in food products than would be obtained using different cultivars of highbush blueberries. Wild lowbush blueberry fruit may also provide “fruitier” and “sweeter” flavors to a food product than would be obtained with cultivated highbush fruit.

Author Contributions: Conceptualization, C.F.F.; methodology, C.F.F. and M.A.J.; supervision, C.F.F.; writing—original draft, C.F.F. and S.Q.; writing—review and editing, C.F.F.; funding acquisition, C.F.F.; investigation, S.Q., M.A.J. and D.M.; formal analysis, M.A.J., D.M. and S.F. All authors have read and agreed to the published version of the manuscript.

Funding: This research was partially funded by the Wild Blueberry Association of North America. Participation in this study by Songshan Qiu was funded by the Chinese Scholarship Council (CSC).

Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki, and approved by the Agriculture and Agri-Food Canada Human Research Ethics Committee (Approval 208-F-001, 24 September 2018).

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Acknowledgments: The authors wish to thank Wilhelmina Kalt for coordinating the acquisition of wild lowbush blueberries used in this study and for proofreading the manuscript; and NovaAgri Inc. for supplying cultivated highbush blueberries used in this study.

Conflicts of Interest: The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

References
1. Forney, C.F.; Kalt, W. Blueberry and cranberry. In Health-Promoting Properties of Fruit and Vegetables; Terry, L.A., Ed.; CABI: Oxfordshire, UK, 2011; pp. 51–73. [CrossRef]
2. Brazelton, C.; Fain, C.; Ogg, M. Global State of the Blueberry Industry Report 2021; International Blueberry Organization: Santiago, Chile, 2022; p. 84.
3. WBANA. Growing Wild Blueberries. Available online: https://www.wildblueberryassociation.ca/the-better-blueberry/the-story-of-wild/growing-wild-blueberries/ (accessed on 9 October 2020).
4. Lobos, G.A.; Hancock, J.F. Breeding blueberries for a changing global environment: A review. Front. Plant. Sci. 2015, 6, 782. [CrossRef] [PubMed]
5. Kalt, W.; Ryan, D.A.J.; Duy, J.C.; Prior, R.L.; Ehlenfeldt, M.K.; Vander Kloet, S.P. Interspecific variation in anthocyanins, phenolics, and antioxidant capacity among genotypes of highbush and lowbush blueberries (Vaccinium section cyanococcus spp.). J. Agric. Food Chem. 2001, 49, 4761–4767. [CrossRef] [PubMed]
6. AgMRC. Blueberries. Available online: https://www.agmrc.org/commodities-products/fruits/blueberries (accessed on 3 March 2022).
7. Gilbert, J.L.; Olmstead, J.W.; Colquhoun, T.A.; Levin, L.A.; Clark, D.G.; Moskowitz, H.R. Consumer-assisted selection of blueberry fruit quality traits. *HortScience* 2014, 49, 864–873. [CrossRef]

8. Forney, C.F.; Kalt, W.; Jordan, M.A.; Vinquyst-Tymchuk, M.R.; Fillmore, S.A.E. Blueberry and cranberry fruit chemistry during development. *J. Berry Res.* 2012, 2, 169–177. [CrossRef]

9. Sater, H.M.; Bizzio, L.N.; Tieman, D.M.; Muñoz, P.D. A review of the fruit volatiles found in blueberry and other Vaccinium species. *J. Agric. Food Chem.* 2020, 68, 5777–5786. [CrossRef]

10. Du, X.; Rouseff, R. Aroma active volatiles in four southern highbush blueberry cultivars determined by gas chromatography-olfactometry (gc-o) and gas chromatography-mass spectrometry (gc-ms). *J. Agric. Food Chem.* 2014, 62, 4537–4543. [CrossRef]

11. Forney, C.F.; Qiu, S.; Jordan, M.A.; Munro Pennell, K.; Fillmore, S. Impact of species, growing location and cultivar on flavor chemistry of blueberry fruit. *Acta Hort.*, 2022, in press.

12. Farneti, B.; Khomenko, I.; Grisenti, M.; Ajelli, M.; Betta, E.; Algarra, A.A.; Cappellin, L.; Aprea, E.; Gasperi, F.; Biasiol, F.; et al. Exploring blueberry aroma complexity by chromatographic and direct-injection spectrometric techniques. *Front. Plant. Sci.* 2017, 8, 617. [CrossRef]

13. Forney, C.F. Horticultural and other factors affecting aroma volatile composition of small fruit. *HortTechnology* 2001, 11, 529–538. [CrossRef]

14. Du, X.; Plotto, A.; Song, M.; Olmstead, J.; Rouseff, R. Volatile composition of four southern highbush blueberry cultivars and effect of growing location and harvest date. *J. Agric. Food Chem.* 2011, 59, 8347–8357. [CrossRef]

15. Beaulieu, J.C.; Stein-Chisholm, R.E.; Boykin, D.L. Qualitative analysis of volatiles in rabbiteye blueberry cultivars at various maturities using rapid solid-phase microextraction. *J. Am. Soc. Hortic. Sci.* 2014, 139, 167–177. [CrossRef]

16. Horvat, R.J.; Schlotzhauer, W.S.; Chortyk, O.T.; Nottingham, S.F.; Payne, J.A. Comparison of volatile compounds from rabbiteye blueberry (Vaccinium ashei) and deerberry (V. Stamineum) during maturation. *J. Essent. Oil Res.* 1996, 8, 645–648. [CrossRef]

17. Partlment, T.H.; Kolor, M.G. Identification of the major volatile components of blueberry. *J. Food Sci.* 1975, 40, 762–763. [CrossRef]

18. Gilbert, J.L.; Guthart, M.J.; Gezan, S.A.; De Carvalho, M.P.; Schwieterman, M.L.; Colquhoun, T.A.; Bartoshuk, L.M.; Sims, C.A.; Clark, D.G.; Olmstead, J.W. Identifying breeding priorities for blueberry flavor using biochemical, sensory, and genotype by environment analyses. *PLoS ONE* 2015, 10, e0138494. [CrossRef] [PubMed]

19. Cheng, K.; Peng, B.; Yuan, F. Volatile composition of eight blueberry cultivars and their relationship with sensory attributes. *Flavor Fragr. J.* 2020, 35, 443–453. [CrossRef]

20. Qian, Y.L.; Zhang, D.; An, Y.; Zhou, Q.; Qian, M.C. Characterization of aroma-active compounds in northern highbush blueberries “bluecrop” (Vaccinium corymbosum “bluecrop”) and “elliott” (V. corymbosum “elliott”) by gas chromatography-olfactometry dilution analysis and odor activity value. *J. Agric. Food Chem.* 2021, 69, 5691–5701. [CrossRef]

21. Cai, X.; Mai, R.Z.; Zou, J.J.; Zhang, H.Y.; Zeng, X.L.; Zheng, R.R.; Wang, C.Y. Analysis of aroma-active compounds in three sweet osmanthus (osmanthus fragrans) cultivars by gc-olfactometry and gc-ms. *J. Zhejiang Univ. Sci B* 2014, 15, 638–648. [CrossRef]

22. Campo, E.; Ferreira, V.; Escudero, A.; Cacho, J. Prediction of the wine sensory properties related to grape variety from dynamic-headspace gas chromatography-olfactometry data. *J. Agric. Food Chem.* 2005, 53, 5682–5690. [CrossRef]

23. Xiao, Z.; Chen, H.; Niu, Y.; Zhu, J. Characterization of the aroma-active compounds in banana (Musa AAA red green) and their contributions to the enhancement of sweetness perception. *J. Agric. Food Chem.* 2021, 69, 15301–15313. [CrossRef]

24. De-La-fuente-blanco, A.; Ferreira, V. Gas chromatography olfactometry (gc-o) for the (semi)quantitative screening of wine aroma. *Foods* 2020, 9, 1892. [CrossRef]

25. Xu, F.; Liu, S.; Liu, Y.; Wang, S. Effect of mechanical vibration on postharvest quality and volatile compounds of blueberry fruit. *Food Chem.* 2021, 349, 129216. [CrossRef] [PubMed]

26. Lugemwa, F.N.; Lwande, W.; Bentley, M.D.; Mendel, M.J.; Alford, A.R. Volatiles of wild blueberry, Vaccinium angustifolium: Possible attractants for the blueberry maggot fruit fly, rhagoletis mendax. *J. Agric. Food Chem.* 1999, 37, 232–233. [CrossRef]

27. Forney, C.F.; Kalt, W.; Vander Kloet, S.P. Comparison of berry composition of selected Vaccinium species (ericaceae) with gaylussacia dumosa. *Botany* 2012, 90, 355–363. [CrossRef]

28. Du, X.; Olmstead, J.; Rouseff, R. Comparison of fast gas chromatography-surface acoustic wave (fgc-saw) detection and gc-ms for characterizing blueberry cultivars and maturity. *J. Agric. Food Chem.* 2012, 60, 5099–5106. [CrossRef] [PubMed]

29. Pico, J.; Gerbrandt, E.M.; Castellarin, S.D. Optimization and validation of a spme-gc/ms method for the determination of volatile compounds, including enantiomeric analysis, in northern highbush blueberries (Vaccinium corymbosum l.). *Foods* 2022, 368, 130812. [CrossRef]

30. Cometto-Muñiz, J.E.; Cain, W.S.; Abraham, M.H.; Kumarsingh, R. Sensory properties of selected terpenes. Thresholds for odor, nasal pungency, nasal localization, and eye irritation. *Ann. N. Y. Acad. Sci.* 1998, 855, 648–651. [CrossRef]

31. Elsharif, S.A.; Buettner, A. Structure-odor relationship study of c-6 unsaturated acyclic monoterpenic alcohols: A comparative approach. *Flavour Sci*; Siegmund, B., Leitner, E., Eds.; Verlag der Technischen Universität Graz: Graz, Austria, 2018. [CrossRef]

32. Hirvi, T.; Honkanen, E. The aroma of blueberries. *J. Sci. Food Agric.* 1983, 34, 992–996. [CrossRef]

33. Qian, Y.P.; Zhou, Q.; Magana, A.A.; Qian, M.C. Comparative study of volatile composition of major northern highbush blueberry (Vaccinium corymbosum) varieties. *J. Food Compos. Anal.* 2022, 110, 104538. [CrossRef]

34. Di Cesare, L.F.; Nani, R.; Proietti, M.; Giombelli, R. Volatile composition of the fruit and juice of some blueberry cultivars grown in Italy. *Ind. Aliment.* 1999, 38, 277–282.
35. Farneti, B.; Emanuelli, F.; Khomenko, I.; Ajelli, M.; Biasioli, F.; Giongo, L. Development of a novel phenotypic roadmap to improve blueberry quality and storability. *Front. Plant. Sci.* **2020**, *11*, 1140. [CrossRef]

36. Ferrão, L.F.V.; Sater, H.; Lyrene, P.; Amadeu, R.R.; Sims, C.A.; Tieman, D.M.; Munoz, P.R. Terpene volatiles mediates the chemical basis of blueberry aroma and consumer acceptability. *Food Res. Int.* **2022**, *158*, 111468. [CrossRef]

37. Horvat, R.J.; Senter, S.D. Comparison of the volatile constituents from rabbiteye blueberries (vaccinium ashei) during ripening. *J. Food Sci.* **1985**, *50*, 429–431. [CrossRef]

38. Song, H.; Liu, J. Gc-o-ms technique and its applications in food flavor analysis. *Food Res. Int.* **2018**, *114*, 187–198. [CrossRef] [PubMed]

39. Rubico, S.M.; McDaniel, M.R. Sensory evaluation of acids by free-choice profiling. *Chem. Senses* **1992**, *17*, 273–289. [CrossRef]