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Investigation of the Aroma Profile of Green Tea Leaves Using Organic Synthesis and Conventional Analytical Techniques

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Abstract: Although many aroma components have been identified in green tea leaves, the aroma compounds contributing to green tea's characteristic odor have not yet been reported. The authors recently reported that aroma components with a matcha-like odor are present in both green tea and black tea prepared from the Sayamakaori tea cultivar. This matcha-like odor is similar to the odor of commercial available matcha (high-quality powdered green tea), and is a specific odor feature of green tea leaves. At present, the green-tea odor is thought to arise from the combination of a large number of constituents. Recent reports indicate that a complex interaction between olfactory receptors and odorants is important for the evaluation of the odors. Taking into consideration these findings, the authors investigated the aroma profile of green tea, focusing on the characteristic molecular structures of the constituents that give matcha-like odor. Using a combination of organic synthesis and gas chromatography-mass spectrometry plus gas chromatography-olfactometry, the authors identified aroma components with matcha-like odors in five other tea cultivars. This investigation also revealed that several compounds with a formyl group were important constituents of the aroma of green tea leaves, although the odor of each constituent was not individually similar to the tea's overall aroma. The authors found for the first time a group of key components that have the matcha-like odor.

Key words: Aroma profile, green tea leaves, matcha-like odor, formyl group, aldehyde, complex odor.

1. Introduction

More than 600 odor constituents have been reported for green tea leaves [1]. The vast majority of these odor constituents and their odor characteristics have been detected by gas chromatography-mass spectrometry (GC-MS) and GC-olfactometry (GC-O) [2]. GC-MS is one of the most effective methods for analyzing volatile components in a complex mixture. However, important constituents in the material sometimes cannot be identified because their concentration is below the detection limit of the technique. In this case, GC-O is a better method for detecting odor constituents in mixtures because the human olfactory system is often more sensitive than GC-MS analysis. A combination of GC-MS and GC-O can be used to evaluate the odor contribution of trace compounds in materials. Many odor compounds from green tea leaves have been detected using these methods and their odor contribution evaluated. For example, linalool and geraniol contribute to the floral odor of green tea, (2*E*)-hexenol and (3*Z*)-hexenol impart a green odor, and dimethylsulfide and 2,5-dimethylpyrazine give a roasted odor. However, no individual odor constituents

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have been reported with a green tea-like odor. The odor of green tea is thought to arise from the combination of a large number of constituents.

In the field of odor science, aroma research has generally focused on individual components in materials [3], and the odors of materials are regarded as a simple combination of these components. However, in some cases, the odor of the material differs from the sum of the odors of the individual compounds [4]. Therefore, instead of analyzing simple combinations of individual odors, analyzing the aroma profiles of materials based on the mechanism of human olfactory reception should be a more ideal approach for the clarification of complex odor. The mechanism of the interaction between odor constituents and human olfactory receptors was unexplained for a long time until Axel and Back published their seminal reports. Recently, Touhara and coworkers [5-7] showed that odor constituents with a similar molecular structure interact with the same olfactory receptor but with different intensities, and that an odor constituent interacts with many different olfactory receptors. Thus, the relationship between the odor constituents and human odor receptors is far more complex than a simple one-to-one interaction. These complex interactions in the olfactory system allow humans to perceive a wide variety of odors. Taking into consideration these recent developments in the understanding of the olfactory reception mechanism, the authors investigated the aroma profile of green tea. They focused on the characteristic molecular structures of the odor constituents and found they consist of many aliphatic compounds and terpenes with similar molecular structures, in addition to nitrogen- and sulfur-containing compounds (Fig. 1). Thus, it could be anticipated that there would be complex odor interactions between the olfactory receptors and odor constituents, and that these interactions would be important for the odor of green tea.

2. Materials and Methods

2.1 Experimental Set Up

All dehydrated solvents and reagents used for synthesis were purchased from Wako Pure Chemical Industries, Ltd., Japan. The 400 MHz ^{1}H NMR and 100 MHz ^{13}C NMR spectra were measured on an Avance 400 NMR system (Bruker, Karlsruhe, Germany). The 500 MHz ^{1}H NMR and 125 MHz ^{13}C NMR spectra were measured on an Avance 500 system (Bruker). Chemical shifts are expressed in ppm relative to the signal of tetramethylsilane as an internal standard (^{1}H and ^{13}C data). Silica gel 60 (Merck, Darmstadt, Germany) was used for column chromatography, silica gel 60 PF254 was used for thin-layer chromatography,

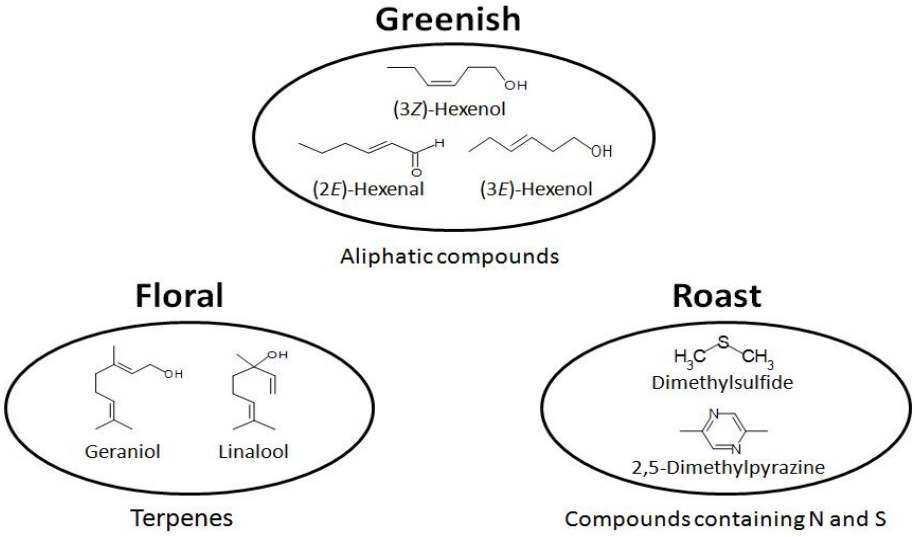

Fig. 1 An overview of green tea constituents.

and silica gel 60 GF was used for preparative thin-layer chromatography (PTLC). The solvent was distilled before use. High-performance liquid chromatography (HPLC) analysis was performed on a LC-9101 (Japan Analytical Industry, Tokyo, Japan) equipped with a UV detector (220 nm). A Shodex 5SIL-10E column (neutral silica gel, Shodex, Tokyo, Japan; 250 mm × 10 mm i.d.; particle size: 5 μm) was used as the normal phase columns. HPLC analysis of a mixture of benzoates was performed using the following conditions: flow rate, 2.0 mL/min; eluent, hexane/isopropyl alcohol = 99.99:0.01 (v/v). GC analysis was conducted with a GC-353 gas chromatography system and OP 275 GC-O analysis system (GL Sciences Inc., Tokyo, Japan) and a HP6890 gas chromatography system (Hewlett-Packard, Wilimington, United States). MS was performed on a JMS-700 AM spectrometer (JEOL, Tokyo, Japan) with electron impact ionization (70 eV). The column was an InertCap Pure-WAX (GL Sciences Inc., 30 m × 0.25 mm i.d.) with a film thickness of 0.25 μm. The column temperature program was as follows: 40 °C (5 min) to 250 °C (30 min) at rate of 4 °C/min. Helium was used as carrier gas at flow rate of 1 mL/min, and the split ratio was 1:30 or splitless. The injection temperature was 250 °C. The retention indices were calculated using a series of n-alkanes (C14, C15, C16, and C20).

2.2 Plant Materials

The tea leaves of *Camellia sinensis* L. cultivars Yabukita and Sayamakaori (green teas), as well as hojicha (roasted tea) were collected at a tea plantation at the Green Tea Laboratory, Saitama Prefectural Agriculture and Forestry Research Center, Japan. The cultivars Samidori and Ujimidori (crude green teas) were collected at a tea plantation at the Green Tea Laboratory, Kyoto Prefectural Agriculture, Forestry and Fisheries Technology Center Tea Industry Research Division, Japan. Commercially available matcha (high-quality powdered green tea) was obtained from Marukyu Koyamaen Co., Ltd., Japan.

2.3 General Procedure for Hexane Extraction

Tea (20-100 g) was added to flasks with hexane (1 L), and stirred at room temperature for 24 h. Removal of the solvent under reduced pressure afforded an oily residue. Extractabilities: 0.56% (Sayamakaori), 1.1% (Yabukita), 1.3% (Samidori), 1.1% (Ujimidori), 4.2% (matcha) and 0.99% (hojicha).

2.4 Fractional Distillation of Hexane Extracts of Green Tea Leaves

The hexane extracts from green tea leaves were fractionally distilled under reduced pressure (96-135 °C, 0.08-0.6 Torr) to afford oily residues. The fraction obtained at lower boiling temperatures had a floral, sweet odor. The fraction obtained at high boiling temperatures had a matcha-like odor.

2.5 Purification of Key Constituents from the Residue with Matcha-Like Odor

The hexane extracts with a matcha-like odor (329 mg) were initially separated by bulb-to-bulb distillation under reduced pressure (96-135 °C, 0.08-0.6 Torr) to afford a green oily residue with a matcha-like odor (193 mg). The residue was separated by column chromatography (hexane/isopropyl alcohol 9:1) to afford four fractions (Fr 1-1: 2.0 mg, Fr 1-2: 64 mg, Fr 1-3: 5.1 mg and Fr 1-4: 5.5 mg). One of the fractions (Fr 1-2: 64 mg) with a matcha-like odor was separated by column chromatography (hexane/chloroform/ethyl acetate) to afford five fractions (Fr 2-1: 71 mg, Fr 2-2: 2.4 mg, Fr 2-3: 6.1 mg, Fr 2-4: 5.5 mg, Fr 2-5: 61.6 mg). One of these fractions (Fr 2-1: 71 mg) with a matcha-like odor was separated by PTLC (chloroform) to afford six fractions (Fr 3-1: 0.5 mg, Fr 3-2: 0.4 mg, Fr 3-3: 1.2 mg, Fr 3-4: 0.2 mg, Fr 3-5: 2.2 mg and Fr 3-6: 57 mg). One of these fractions (Fr 3-6: 57 mg) with a matcha-like odor was separated by PTLC (chloroform/tetrachloromethane 9:1) to afford 11 fractions without a matcha-like odor (Fr 4-1: 0.1 mg, Fr

4-2: 0.2 mg, Fr 4-3: 0.1 mg, Fr 4-4: 10.2 mg, Fr 4-5: 9.6 mg, Fr 4-6: 3.4 mg, Fr 4-7: 0.6 mg, Fr 4-8: 0.2 mg, Fr 4-9: 0.4 mg, Fr 4-10: 0.8 mg and Fr 4-11: 3.6 mg).

2.6 General Procedure for Reducing the Oil with a Matcha-Like Odor

$LiAlH_4$ (70.3 mg) and absolute diethyl ether (3.0 mL) were added to a flask purged with nitrogen. A solution of the green tea residue (62.6 mg) in ether (4.0 mL) was added to the flask. The solution was stirred for 2.5 h at 23 °C. The reaction was monitored by TLC (SiO_2, CH_2Cl_2). Diethyl ether (2.0 mL), water (0.70 mL), 15% NaOH aq. (0.70 mL) and water (2.1 mL) were added slowly to the reaction mixture. The organic layer was separated, washed with saturated aqueous ammonium chloride (2.0 mL × 2) and brine (2.0 mL × 2) and dried over magnesium sulfate. Removal of the solvent gave the crude compounds as greenish oil (54.4 mg). The reduction products (54.4 mg) were purified by PTLC (hexane/isopropyl alcohol 95:5) to afford a mixture of alcohols (5.70 mg).

2.7 Synthesis of Benzoate Derivatives of Alcohols Obtained from Reduction of the Residue

The mixture of alcohols (5.70 mg) was dissolved in absolute diethyl ether (1.0 mL). Absolute pyridine (10.2 mg, 0.13 mmol) and benzoyl chloride (15.5 mg, 0.11 mmol) were added to the solution and stirred for 3 h. The progress of the reaction was monitored by TLC (SiO_2, CH_2Cl_2). Diethyl ether (1.0 mL) and water (0.5 mL) were added to the reaction mixture. The reaction mixture was washed with water (0.5 mL × 3) and brine (0.5 mL × 3). The organic layer was dried over anhydrous magnesium sulfate. Removal of the solvent gave a mixture of benzoates as crude oils. The mixture was separated by HPLC to afford the benzoates (0.40 mg).

Benzoates (C7-14) colorless liquid; ^{1}H-NMR (500 MHz, $CDCl_3$) δ7.55 (t, 1H), 7.44 (t, 2H), 8.04 (dd, 2H), 4.31 (t, 2H), 1.77 (m, 2H), 1.25-1.44 (m, 8-22H), 0.88 (m, 3H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ128.3, 129.5, 130.6, 132.8, 166.7, 65.1 (-OCO*C*H_2-), 28.7 (-OCOCH$_2$*C*H$_2$-), 26.0 (-OCOCH$_2$CH$_2$*C*H$_2$-), 29.2, 29.4, 29.5, 29.6, 29.7 [(-CH_2-)$_{1-8}$], 31.9 (-*C*H$_2$CH$_2$CH$_3$), 22.7 (-CH$_2$*C*H$_2$CH$_3$), 14.1 (-CH$_2$CH$_2$*C*H$_3$).

2.8 Synthesis of Aliphatic Aldehydes (C8, C10)

Pyridinium chlorochromate (654 mg, 3.0 mmol), distilled dichloromethane (4.0 mL) and 1-decanol (324 mg, 2.0 mmol) in CH_2Cl_2 (1 mL) were added to a flask purged with nitrogen. The solution was stirred for 1 h at room temperature. The reaction was monitored by TLC (SiO_2, CH_2Cl_2). Diethyl ether (4.0 mL) was added to reaction mixture, and the solid was filtered through Florisil and washed with diethyl ether (2.5 mL × 3). Removal of the solvent gave a crude oil. The mixture was purified by bulb-to-bulb distillation (44-47 °C at 2.0 Torr) to afford 1-decanal (1) (12.2 mg, 3.8%) as a colorless oil. Using a similar procedure, 1-octanal (2) (25.6 mg, 5.1%) was synthesized from 1-octanol (2) (500 mg, 3.84 mmol).

1-Decanal (1) colorless liquid; ^{1}H-NMR (500 MHz, $CDCl_3$) δ0.88 (t, 3H), 1.27-1.30 (m, 12H), 1.60-1.67 (m, 2H), 2.40-2.44 (td, 2H), 9.77 (t, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ14.1 (C10), 22.1 (C3), 22.6 (C9), 29.1 (C6), 29.2 (C7), 29.3 (C4), 29.4 (C5), 31.8 (C8), 43.9 (C2), 203.0 (-CHO).

1-Octanal (2) colorless liquid; ^{1}H-NMR (500 MHz, $CDCl_3$) δ0.89 (t, 3H), 1.23-1.36 (m, 8H), 1.62-1.65 (m, 2H), 2.41-2.44 (td, 2H), 9.77 (t, 1H); ^{13}C-NMR (125 MHz, $CDCl_3$) δ14.1 (C8), 22.2 (C3), 22.7 (C7), 29.1 (C5), 29.2 (C4), 31.8 (C6), 44.0 (C7), 203.1 (-CHO).

3. Results and Discussion

In previous studies, the authors have investigated the complex odor of several materials considering the mechanism of human olfactory reception. For example, it was found that the complex odor of frankincense arose from three groups of compounds that contained odor constituents with similar structures [8] (Fig. 2a), and that the group with frankincense-like odor was composed of incensole derivatives having similar

structures. Here, the same approach has been used to clarify the odor of green tea via the bulb-to-bulb distillation of the hexane extracts (Fig. 2b).

Initially, the authors studied the odors of green and black tea leaves [9] because their odors are markedly different, even though they are made from the same plant (*Camellia sinensis* L.). When the black tea from the green tea cultivar Sayamakaori was prepared by the conventional method, the aroma of the prepared black tea leaves differed from that of commercial black tea leaves. The authors focused on the difference between the aroma profiles of Sayamakaori black tea and commercial black tea. The tea leaves were extracted with hexane, and the hexane extracts were separated into distilled oil and residue by bulb-to-bulb distillation. Only the residue from commercial black tea was odorless; the residues from the green and black Sayamakaori teas had a matcha-like odor. This indicates that the difference in the odors of the residues caused the difference in the odors of the two types of black tea (Table 1). The authors defined matcha-like odor as the specific odor feature of green tea. In Japan, the number of foods with green tea-like odor is almost equal to the number of foods containing commercially

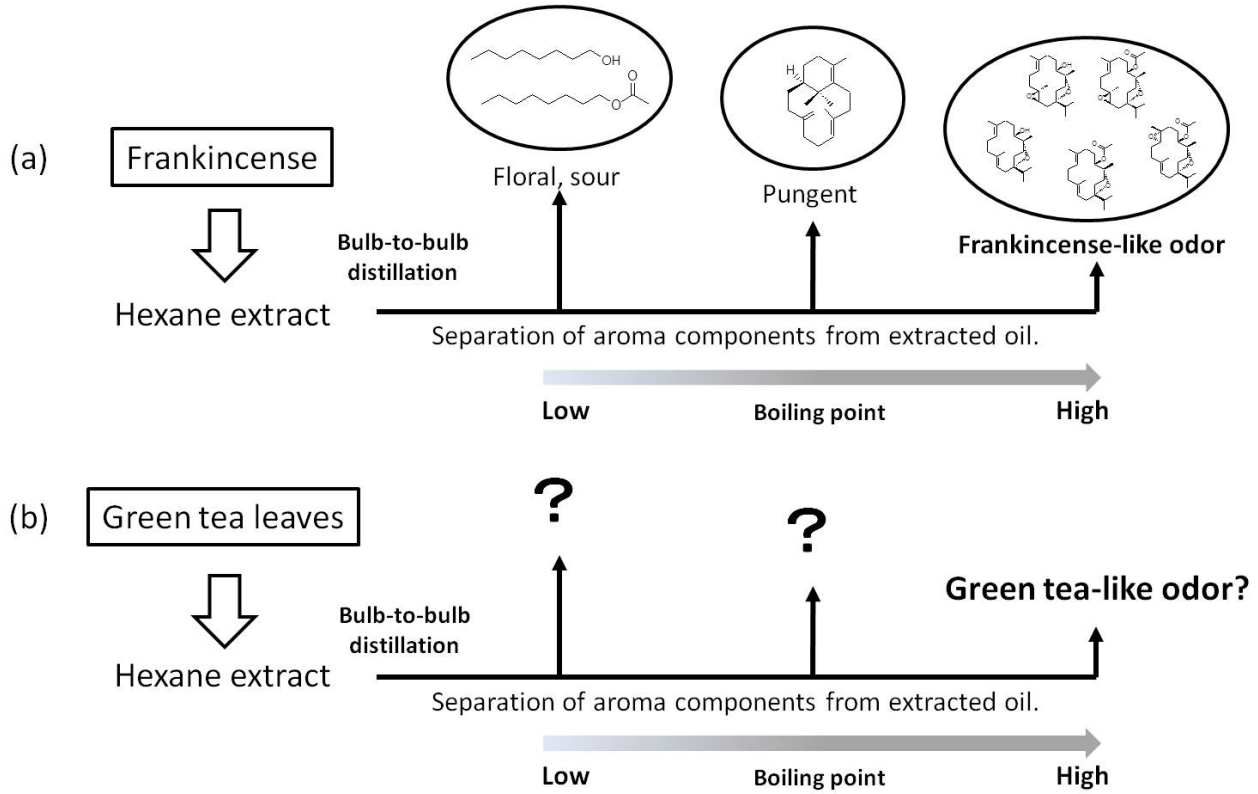

Fig. 2 Method for grouping constituents by bulb-to-bulb distillation: (a) previous results for frankincense; (b) method used for green tea leaves.

Table 1 Comparison of the aroma profiles of green tea leaves and black tea leaves derived from the Sayamakaori cultivar.

Material (odor)	Odor of distilled oil (°C/Torr)	Odor of residue
Green tea (fresh, hay-like, slightly sweet)	Floral, sweet, bitter (75-112/0.10)	Matcha-like
Black tea[a] (hay-like, floral)	Floral, sweet (54-93/0.20)	Matcha-like
Black tea[b] (floral)	Floral, sweet (63-131/0.10)	Odorless

[a] Prepared from tea cultivar Sayamakaori.

[b] Commercially available Darjeeling tea (Twinings of London).

available matcha. The odor of matcha is similar to that of green tea, and was revealed to contribute to the specific odor character of green tea leaves.

These results suggest that the matcha-like odor is characteristic of green tea leaves.

The authors investigated whether the matcha-like odor is the common base odor in all green tea leaves [10]. They used the following six types of green tea leaves. First, four different green tea cultivars were investigated: Yabukita, Sayamakaori, Ujimidori, Samidori (Table 2). Yabukita and Sayamakaori are standard tea cultivars in Japan. Ujimidori and Samidori are produced in Kyoto, Japan, and have odor features distinct from those of "Yabukita" and "Sayamakaori". All four cultivars are used to produce sencha, which is the most common type of Japanese green tea.

Second, teas obtained from two other processing methods, hojicha and matcha, were also investigated (Table 3). Hojicha is obtained by roasting green tea and matcha is high quality powdered green tea obtained from tencha, specially processed tea leaves. Hojicha and matcha have their own characteristic odors, which are different from that of sencha and are a result of their distinct processing methods.

The green tea leaves were extracted with hexane at room temperature, and hexane extracts were divided into the distilled oil and the residue by bulb-to-bulb distillation (Fig. 2b). The distilled oils had different odors depending on the tea cultivars and processing methods. However, the matcha-like odor was present in all the tea leaves except for those of matcha (Table 4). GC-O analysis was performed on the matcha residue to investigate the differences. The authors identified a group of compounds that produced a matcha-like odor, but also detected the peaks giving the strong laver-like odor in the residue. They presumed that this reduced the prominence of the matcha-like odor.

The results show that the matcha-like odor is a base note for green tea leaves. To identify the components of the matcha-like odor, NMR analysis was performed on all the residues obtained from the extracts. The large number of peaks in the 8-10 ppm region indicated the

Table 2 Odors of green teas from different cultivars.

Cultivar	Yabukita	Sayamakaori	Ujimidori	Samidori
Odor	Sencha-like	Sencha-like astringent	Shaded aroma (weak)	Shaded aroma (strong)

Table 3 Odors of green teas obtained by different processing methods.

Manufacture	Green tea[a]	Hojicha[a]	Matcha[b]
Odor	Sencha-like	Roast, toasty	Laver

[a] Prepared from tea cultivar Yabukita.

[b] Commercially available tea from Marukyu Koyamaen Co., Ltd., Japan.

Table 4 Separation of the distilled fraction from the hexane extracts of green tea leaves by precise bulb-to-bulb distillation.

Manufacture	Cultivar	Odor of distilled oil (°C/Torr)	Odor of residue
Crude tea[a]	Yabukita	Bitter, sweet (96-130/0.1-0.15)	Matcha-like
Crude tea[a]	Sayamakaori	Roast, sweet (97-134/0.1-0.4)	Matcha-like
Crude tea[a]	Samidori	Flavoursome, burnt (97-138/0.15-2.0)	Matcha-like
Crude tea[a]	Ujimidori	Flavoursome, burnt (97-134/0.4-0.6)	Matcha-like
Hojicha	Yabukita	Hojicha-like, burnt (97-134/0.4-0.6)	Matcha-like
Matcha	-	Roast (82-127/0.6-1.0)	Laver

[a] Crude tea consists of green tea leaves, to later be made into sencha.

presence of many compounds with a formyl group (Fig. 3).

The authors focused on aldehydes as the key compounds for the matcha-like odor of green tea leaves because aldehydes are common key compounds in the fragrance and flavor industries [3], and many aldehydes have been reported as odor constituents of green tea leaves [1].

The residue was divided into several fractions by column chromatography and PTLC of the residue of the extracts. Some fractions with a matcha-like odor contained aldehydes as the main constituents. The fractions were repeatedly purified to isolate the individual aldehyde components and it was found that the matcha-like odor decreased during the purification process. These results indicate that certain aldehydes are key compounds for the matcha-like odor. Next, a synthetic study was performed to confirm that the aldehydes are the key compounds. Aldehydes are easily reduced to their corresponding alcohols. Generally, alcohols are more stable than aldehydes, although their odor is weaker. The authors focused on the change of odor before and after the reduction of the residue. After the $LiAlH_4$ reduction, the matcha-like odor was lost, coinciding with the disappearance of several aldehydes (Fig. 4).

These results suggest that a group of aldehydes are the key components of the matcha-like odor. The authors attempted to isolate these alcohols to elucidate the structure of the corresponding aldehydes. However, the purification was difficult because the alcohols had very similar structures. On the other hand, the benzoate derivatives of the alcohols could be more easily separated. The structures of several benzoates were investigated by 1D and 2D NMR. The six characteristic peaks (65.1, 31.9, 28.7, 26.0, 22.7 and 14.1 ppm) in the ^{13}C-NMR spectra showed that the benzoates contain aliphatic chains longer than C6 chain. Moreover, the 2D C,H-COSY spectra and the peak intensity in the 1D ^{13}C-NMR indicated that the benzoates were a mixture of C7-14 hydrocarbon chain derivatives. These results show that aldehydes with a matcha-like odor possessed C7-14 hydrocarbon chain moieties, which were reported in a previous paper to be important constituents of the green tea odor [1]. The authors synthesized two of the aldehydes, 1-octanal and 1-decanal, to verify whether these compounds were present in the residue.

However, it was unable to confirm the presence of 1-octanal and 1-decanal in the residue despite attempting to detect these compounds by NMR. The aldehyde content of the extract may be decreased by

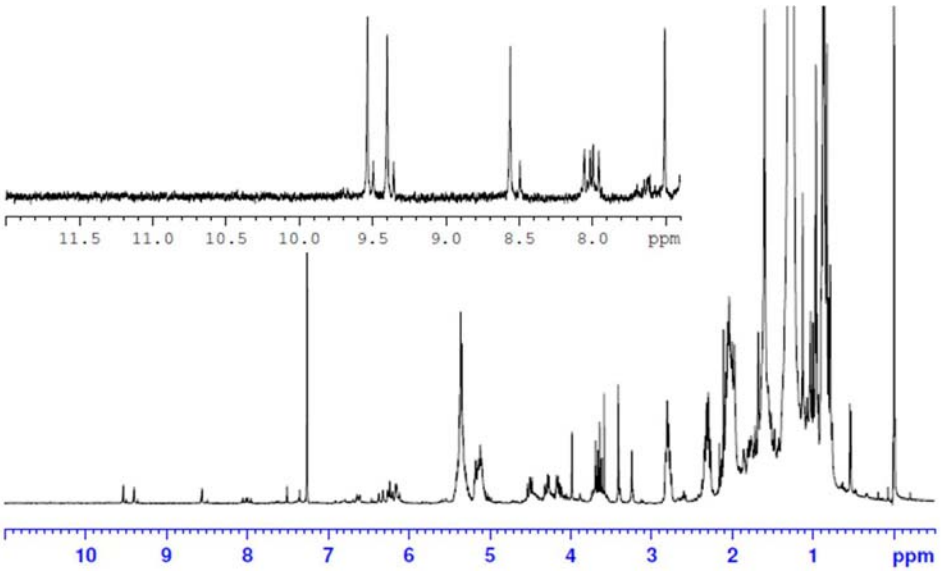

Fig. 3 ^{1}H NMR spectra (500 MHz, $CDCl_3$) of the residue obtained from the hexane extracts of the Sayamakaori cultivar.

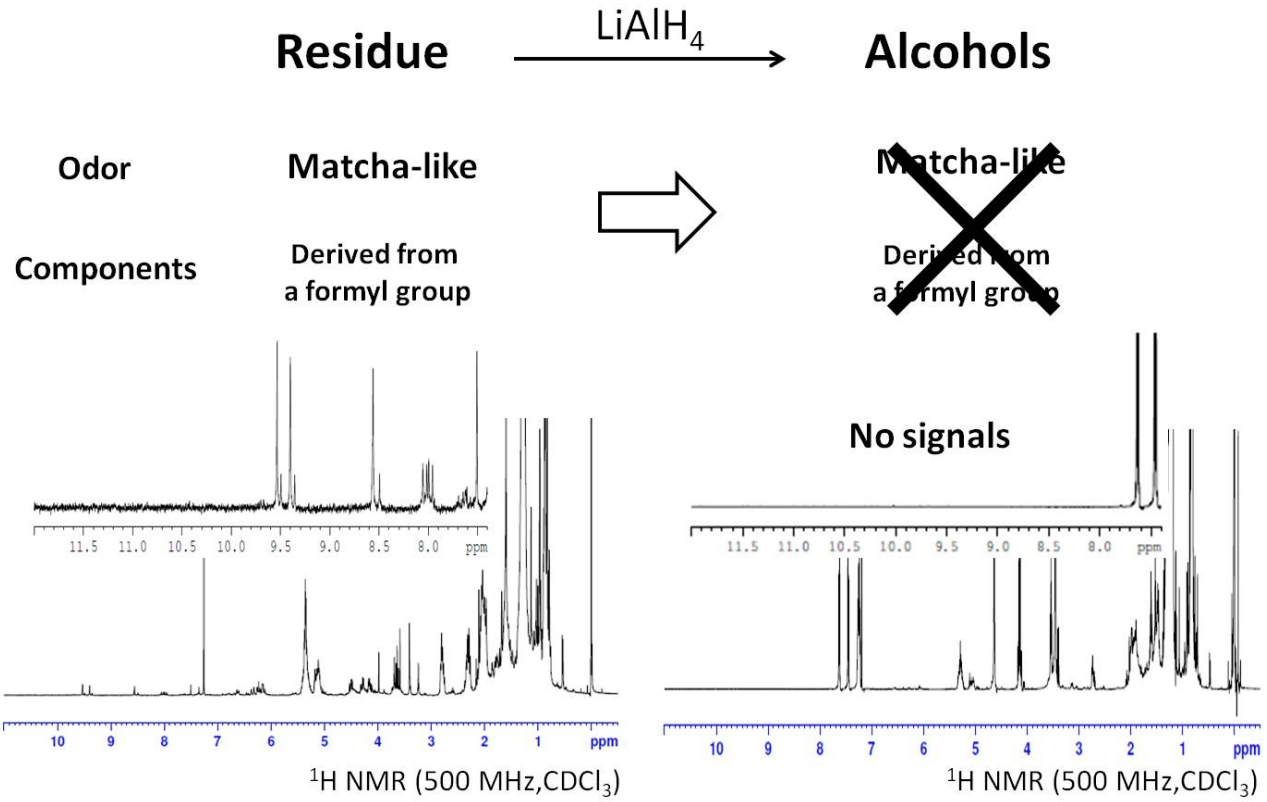

Fig. 4 Change of ^{1}H NMR spectra (500 MHz, $CDCl_3$) caused by the reduction of the residue.

repeated separation because of the instability of these compounds. Therefore, the authors checked the stability of 1-octanal and 1-decanal. The half-life of the aldehydes was 50-60 h at room temperature in the presence of air. However, the authors believe that many aldehydes are key compounds for the matcha-like odor for the following reasons. The matcha-like odor was lost upon reduction of the residue, and this loss of odor coincided with the disappearance of several aldehydes. During separation, the chromatographic fractions with a matcha-like odor always contained some aldehydes. Furthermore, more than 60 aldehydes have been reported in the literature as odor constituents of green tea leaves [1].

The residue of the tea extracts from the Sayamakaori cultivar was examined by NMR to identify the odor constituents responsible for the matcha-like odor. The high-intensity signals in the 9.4-9.6 ppm aldehyde proton region were not derived from C7-14 hydrocarbon chain aldehydes. The authors determined the structure of the compound with signals in this region. However, the fraction containing this compound as a main constituent was odorless. These results indicate that the compound was not a key component of the matcha-like odor. Nonetheless, the results still suggest that the aldehydes are the key compounds responsible for the matcha-like odor. Next, the authors focused on the minor compounds in the extracts as key constituents of the matcha-like odor. They identified the key constituents by GC-O and GC-MS analysis.

Compounds with a retention time between 25-36 min in the GC-O chromatogram possessed odors similar to the key components, although there were no signals corresponding to the expected key compounds. After 50 min, there were many signals, although these corresponded to odorless compounds. Therefore, the key compounds eluted at 25-36 min. Moreover, many signals with a retention time of 10-70 min disappeared upon reduction, which also coincided with a loss of

odor. This also supports the overall conclusion. Indeed, similar results were obtained for the Yabukita, Ujimidori, Samidori and hojicha tea extracts. To verify these important key constituents, the authors performed GC-MS in addition to GC-O. GC-MS analysis showed that all the high-intensity signals were esters with long chain hydrocarbon chain moieties and hydrocarbons. These compounds are odorless. This was consistent with the GC-O analysis. The compounds with high signal intensities were not key compounds for the matcha-like odor (RT: after 50 min). Next, the authors focused on the minor trace constituents (RT: 10-36 min) and elucidated the structures of these constituents by GC-MS analysis using previously reported GC analytical data in addition to the GC-MS data [11-13] (Table 5).

Table 5 GC-MS analysis of the residue with a matcha-like odor.

Compounds	Running time (min)	Retention index	Peak area[b] (%)
Ethyl 3-Methylcrotonate	12.67	1214	t[c]
Tribromomethane	13.09	1228	t
1-Octanal[a]	14.16	1263	t
Unknown	18.67	1440	t
Unknown	19.17	1434	t
Unknown	20.25	1466	t
1-Nonene	20.75	1481	t
Benzaldehyde[a]	21.93	1516	t
(*E*,*Z*)-2,6-Nonadienal[a]	22.22	1525	t
Unknown	23.42	1562	t
(*E*)-2-Nonenal	24.09	1582	t
1-Nonanal	24.34	1590	t
(*Z*)-2-Octenol[a]	24.37	1591	t
1,2-Benzendicarboxylic acid	24.67	1600	t
1-Methyl-*N*-nitrosodiethylamine	25.17	1619	t
Safranal[a]	25.80	1641	t
Unknown	26.50	1667	t
(*E*,*E*)-2,4-Nonadienal[a]	26.60	1671	t
Unknown	26.84	1680	t
Unknown	27.17	1692	t
1-Decanol	28.75	1745	t
(*E*,*Z*)-2,4-Decadienal[a]	28.91	1750	t
α-(*tert*-Butyl)benzenemethaneimine	29.42	1767	t
2-*tert*-Butyl-1*H*-indole	31.42	1833	0.24
1-Undecanol	32.00	1852	t
1,7-Dimethylnaphthalene 2,3-Dimethylnaphthalene	33.09	1888	t
7-Tetradecene	33.75	1910	t
1-Undecene	35.09	1957	t
4-Pyridinecaboxaldehyde	36.50	2005	t
Ethyl linoleate	38.59	2071	8.5
4-Phenylpyridine	39.00	2084	t
5-Eicosyne	39.42	2097	t
Linoleic acid	40.09	2119	t
Nonanoic acid	41.92	2180	t
1-Furfurylpyrrole	47.42	2361	0.24
3-Hydroxy-2',5'-dichlorobiphenyl	49.92	2443	0.12
Ethyl benzimidate	50.17	2451	t

(Table 5 continued)

Compounds	Running time (min)	Retention index	Peak area[b] (%)
2,3-Dihydro-5-methyl-1-(p-tolyl)-1*H*-benzimidazol-2-one	51.25	2487	0.18
N-[2-(Phenylmethylene)butylidene]cyclohexanamine	51.59	2498	t
Dibenzobenzimidazole	51.84	2507	0.6
6-Octadecenal	52.59	2531	1.4
Methyl 2-amino-3,4,5-trimethoxybenzoate	53.42	2559	10.3
1,1'-Biphenyl-2-amine	54.17	2583	t
2,3-Dimethylbenzoic acid 2,5-Dimethylbenzoic acid	54.42	2592	t
5-Eicosyne	54.59	2597	t
o-[1-(4-hydroxyphenyl)-1-methylethyl]phenol 4-Nitrodiphenylmethane	55.09	2614	t
7-Octadecenal	55.75	2635	t
Octadecanoic acid (*Z*)-9-hexadecenyl ester	57.84	2704	t
10-Octadecynoic acid methyl ester 12-Heptadecyn-1-ol 1,2-Nonadecanediol	60.00	2775	t
6-Octadecenal	61.25	2817	10.2
3-Eicosyne	62.09	2845	1.2
Caffeine	65.75	2966	0.61
Squalene	66.84	3002	5.1
3-Octadecenal	67.17	3013	0.73
Bis(2-ethylhexyl) phthalate	68.75	3064	0.85
11-Hexacosyne	70.09	3109	t
Oleyl alcohol	70.59	3125	20.7
3-Eicosyne	75.75	3295	0.73
6-Benzylquinoline 2-Benzylquinoline	78.67	3392	t
1,2-Nonadecadienol	82.34	3596	36.5
Total	-	-	98.2

[a] These compounds were assigned by using previously reported GC analytical data [11-13].

[b] The area percentage composition was obtained by peak area normalization without considering relative response factors.

[c] $t < 0.1\%$.

It could be found that a series of aliphatic molecules were key compounds for the matcha-like odor: (2*E*)-nonenal, 1-nonanal, 1-decanol, 1-octanal, (2*Z*)-octen-1-ol, (2*E*,4*Z*)-decadienal, (2*E*,4*E*)-nonadienal and (2*E*,6*Z*)-nonadienal.

4. Conclusions

The authors have investigated the odor of green tea using a combination of synthetic organic methods and analytical techniques. A group of key compounds that produces the matcha-like odor as the base note of green tea's distinct odor were newly identified.

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