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ORIGINAL

Retention Indices for Identification of Aroma Compounds by GC: Development and Application of a Retention Index Database

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Abstract In this paper we focus on the development and application of a retention index database for aroma compounds with high structural diversity. Retention indices of over 300 aroma compounds were determined on three capillary columns of different polarity. The capability of the retention index database was evaluated by identification of the components of tobacco flavor. The results showed that the database can be used for qualitative identification. The database also contains a good set of retention index data for study of quantitative structure–retention relationships.

Keywords Gas chromatography · Retention index · Aroma compound · Database · Tobacco flavor analysis

Introduction

Aroma compounds, which have been widely used in food, perfume, medicine, and tobacco, among other products, are economically important. Accurate identification of aroma compounds is a prerequisite for quality control and product development [1–3]. In recent decades, gas chromatography–mass spectrometry (GC–MS) has been proved to be a useful tool for identification of volatile and semi-volatile organic compounds because of its high separation performance and accurate identification capability [4–7]. The main purpose of GC–MS analysis is to resolve mixtures of compounds into less complex mixtures or, ultimately, pure components. Usually, compound identification is achieved by library search on the basis of comparison of experimental mass spectra with those stored in a suitable library (e.g., that of the National Institute of Standards and Technology, NIST) [8]. However, mass spectra cannot provide enough structural information to distinguish all compounds easily. In addition to matching of mass spectra, chromatographic retention data can serve as complementary information for positive identification of resolved components. The retention index (RI), a useful tool for qualitative identification, has been used by many analysts [9–13]. The RI was first proposed by Kováts in 1958 for isothermal chromatography and further developed by van den Dool and Kratz into the linear temperature-programmed retention index (LTPRI) for linear temperature programming. Because it is independent of operating conditions, except for stationary phase polarity [14, 15], the RI is very suitable for interlaboratory comparison and provides a feasible way of investigating potential mechanisms of chromatographic retention behavior. When RI are used for qualitative identification, both homemade RI and published RI databases can be used as reference. This approach avoids the use of time-consuming and expensive procedures in which identification is based on injection of pure compounds, especially for complex samples containing hundreds of components for which the corresponding standard pure compounds may not be available.

Nowadays, linear temperature-programmed GC is used for a wide range of applications, and LTPRI has

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thus become increasingly important for compound identification. Applications of this technique have increased substantially, because of the possibility of converting non-volatile or thermally unstable compounds into volatile and fairly stable derivatives. The use of RI in temperature-programmed GC has been reviewed by Gonzales [16]. Development of LTPRI databases for GC–MS analysis is extremely necessary, especially when mass spectral data are far more readily available than retention index data. In fact, since the early days of GC much effort has been devoted to standardization of the methods used to determine retention data, especially the retention index, with the main purpose being wider and more reliable use of collated published results for identification purposes. Several retention index databases are available [17, 18]. Worthy of note, among several retention index datasets, are compilations published as books and widely used as reference works, for example those authored by Jennings and Shibamoto [19] and by Adams [20]. However, most of the aforementioned index databases are collected from different published studies. Thus, it is worth noting that discrepancies may arise because the retention index data have been reported by many different laboratories and reported operating conditions are not always reliable. Certainly, the best RI database is that developed in a single laboratory with fixed experimental conditions, but this is not an easy task because it is time-consuming and expensive. For aroma compounds, the retention index database at http://www.flavornet.org/f_kovats.html has been widely used. However, the retention indices of this database were collected from many papers in recent decades, and only Kováts RI for isothermal conditions are included. To the best of our knowledge, there are very few comprehensive databases of LTPRI of aroma compounds measured on different stationary phases by a single laboratory. One exception is the study of Federica Bianchi in which an RI database of 250 food aroma volatiles on a polar stationary phase was created [21]. Moreover, only one polar stationary phase was used in that study. In addition, mixtures rather than pure standard compounds were used for measurement of retention indices, and identification of the volatile compounds was performed by comparing mass spectra with those stored in the NIST library. So, there is a problem of qualitative accuracy. In flavor and fragrance analysis, LTPRI in gas chromatographic analysis has been reviewed by Zellner et al. [22].

In this context, the purpose of this study was to develop an LTPRI database of over 300 aroma compounds and to use it for analysis of tobacco flavor. Alcohols, esters, aldehydes, ketones, ethers, organic acids, and phenols, among other compounds, were included in the database. Retention times of these aroma compounds on three capillary columns (HP-1, HP-5MS, and DB-225MS) of different polarity were determined; the C_7–C_{30} *n*-alkanes were used as reference substances for LTPRI calculation. To evaluate the stability of the LTPRI, experiments were performed under different experimental conditions: heating rate (4, 6, 8 or 10 °C min^{-1}), flow rate (1 or 2 mL min^{-1}), and initial temperature (50, 60, 70, 80, or 90 °C). The results showed the LTPRI were stable under the different experimental conditions, differences being <10 retention index units for most compounds. Twenty-two compounds for which differences were >10 retention index units are discussed on the basis of molecular structure. We also studied the relationship between retention index and experimental conditions, and retention index normalization. Finally, the developed LTPRI database was used for analysis of tobacco flavor. The results obtained show that many chemical components could be identified by RI matching using this database.

Experimental

Instruments and Samples

GC–MS analysis was performed with an Agilent (USA) 7890A gas chromatograph equipped with an Agilent 5975C mass spectrometer. Three commonly used stationary phases of different polarity were used:

- HP-5MS capillary column (30 m × 0.25 mm i.d., film thickness 0.25 μm; Agilent);
- HP-1 capillary column (30 m × 0.25 mm i.d., film thickness 0.25 μm; Agilent); and
- DB-225MS capillary column (30 m × 0.25 mm i.d., film thickness 0.25 μm; Agilent).

Helium (99.99 %, Changsha Industrial Gas) was used as carrier gas at a flow rate of 1.0 mL min^{-1}. Split injection of 1 μL was conducted with a split ratio of 15:1. The oven temperature program was: initial temperature 50 °C for 1 min, increased at 6 °C min^{-1} to 280 °C which was maintained for 2 min, the precision of temperature control was ±0.1 °C. Mass spectrometry was performed in electron-impact (EI) mode, scan range 33–350, ionization energy 70 eV. The injector and ion source were kept at 250 and 230 °C, respectively. The MS was set to acquire data in "Raw" and "Scan" modes. The volatile compounds were identified by comparing their mass spectra with those stored in the National Institute of Standards and Technology (NIST), US Government library.

All standard aroma compounds were purchased from Shanghai H&K Flavor. C_7–C_{30} *n*-alkanes were purchased from Sigma–Aldrich, Milan, Italy.

RI Calculation

Retention indices, with C_7–C_{30} *n*-alkanes as reference substances, were calculated by use of the expression:

$$\mathrm{RI}(x) = 100 \times z + 100 \times \frac{\mathrm{RT}(x) - RT(z)}{\mathrm{RT}(z+1) - RT(z)} \quad (1)$$

where RI(x) is the retention index of unknown compound x, z is the number of carbon atoms of the *n*-alkane eluting before unknown compound x, $z + 1$ is the number of carbon atoms of the *n*-alkane eluting after unknown compound x, RT(x) is the retention time of unknown compound x, RT(z) is the retention time of the *n*-alkane eluting before unknown compound x, and RT($z + 1$) is the retention time of the *n*-alkane eluting after unknown compound x. All the indices were calculated for three replicate injections of the pure compounds.

Results and Discussion

Using the experimental conditions given in the section "Instruments and Samples", GC–MS analysis was performed for the aroma compounds and the C_7–C_{30} *n*-alkane reference compounds. On the basis of previous experience, the aroma compounds were divided into 50 groups, i.e. approximately six aroma compounds were injected at a time, to avoid peak overlapping. To assess the reproducibility of retention index values, each aroma compound was analyzed three times under the same experimental conditions. On the basis of the known mass spectra of the compounds analyzed, mass spectral matching was also used for further confirmation of the identity of the aroma compounds. The retention times of the aroma compounds and the C_7–C_{30} reference compounds were then recorded and used to calculate retention indices. As suggested by Ettre, retention indices are reported as integers [23]. Finally, a retention index database of over 300 aroma compounds on three stationary phases was created. Also, taking into account that different GC experimental conditions may be used for identification of aroma compounds in other laboratories, we determined the retention indices under different conditions, e.g. gas flow rate, initial temperature, and heating rate, to evaluate the stability of the retention indices.

Effect of Stationary Phase Polarity

As we know, the mechanism of gas chromatographic retention is interaction between the solute and the stationary phase, thus, RI varies with stationary phase. The effect of stationary phase polarity on retention index has been studied for a long time, and retention index conversion methods have been proposed [24–26]. However, standardization of the retention index remains an unsolved problem, especially conversion of retention index values between non-polar and strongly polar stationary phases. Therefore, the many retention data obtained by use of different stationary phases of different polarity are quite valuable for study of retention index standardization. Data mining studies of quantitative structure–retention relationships (QSRR) for the retention index on different stationary phases have also been reported [27, 28], but this kind of retention index database is quite uncommon, especially for aroma compounds with structural diversity. Much research has revealed large differences between RI values for the same compound obtained on stationary phases of different polarity. Furthermore, because the interaction between analytical solutes and polar stationary phases is more complicated than that between analytical solutes and non-polar stationary phases, the retention index on a polar stationary phase is more difficult to predict [29, 30].

In this paper, three frequently used stationary phases of different polarity were used. HP-1, HP5-MS, and DB-225MS are non-polar, weakly polar, and polar stationary phases, respectively. Retention indices on the three stationary phases are listed in Table 1. The precision of the retention indices was assessed by performing triplicate injections under fixed experimental conditions: differences of <1 retention index unit were observed for all the compounds. As expected, retention indices on the stationary phases HP-1 and HP5-MS were similar, because these two stationary phases are of similar polarity, although retention indices on HP5-MS were always slightly larger than those on HP-1. In contrast, the values of the retention indices on DB-225MS were much larger than those on HP-1 and HP5-MS, because of larger intermolecular forces between the solutes and the polar stationary phase. Interaction between solute and stationary phase depends not only on dispersion force, induction force, and steric effect but also on dipole–dipole interactions, i.e. a directional force on polar stationary phases [29]. Further research on the relationship between molecular structure and differences between RI will be performed in our next study.

A $\triangle$RI of approximately 100 was usually obtained for homologous compounds differing by one carbon atom unit, confirming the reliability of the data [21]. On the basis of this relationship we estimated the reliability of the created RI database. For instance, the retention indices of nonanal, decanal, undecanal, and dodecanal were, respectively, 1,082, 1,183, 1,285 and 1,387 on HP-1, 1,104, 1,205, 1,306 and 1,408 on HP-5MS, and 1,283, 1,394, 1,475 and 1,613 on DB-225MS. We found that the relationship between number of carbon atoms and the corresponding retention index was very significant for HP-1 and HP-5MS but not so obvious for DB-225MS, because factors other than the number of carbon atoms are important in the retention

Table 1 Retention indices of aroma compounds on three columns

CAS	Chemical name	HP-1	HP5-MS	DB-225MS
000102-20-5	Benzeneacetic acid, 2-phenylethyl ester	1865	1922	2709
000112-14-1	Acetic acid, octyl ester	1193	1210	1440
000141-78-6	Ethyl Acetate	*536*	*568*	819
000093-18-5	Naphthalene, 2-ethoxy-	1489	1528	2100
000102-13-6	Benzeneacetic acid, 2-methylpropyl ester	1360	1392	1829
001504-74-1	2-Propenal, 3-(2-methoxyphenyl)-	1480	1533	2333
000106-32-1	Octanoic acid, ethyl ester	1179	1196	1410
000097-62-1	Propanoic acid, 2-methyl-, ethyl ester	744	757	934
000695-06-7	2(3*H*)-Furanone, 5-ethyldihydro-	1005	1054	1710
000112-31-2	Decanal	1183	1205	1494
000488-10-8	2-Cyclopenten-1-one, 3-methyl-2-(2-pentenyl)-, (*Z*)-	1364	1401	1938
000110-19-0	Isobutyl acetate	757	773	970
002721-22-4	2*H*-Pyran-2-one, tetrahydro-6-nonyl-	1874	1926	2753
000123-69-3	Oxacycloheptadec-8-en-2-one, (8*Z*)	1901	1938	2462
000556-82-1	2-Buten-1-ol, 3-methyl-	758	776	1113
000106-29-6	Butanoic acid, 3,7-dimethyl-2,6-octadienyl ester, (*E*)-	1536	1560	1854
005471-51-2	2-Butanone, 4-(4-hydroxyphenyl)-	1498	1553	2675
000104-54-1	2-Propen-1-ol, 3-phenyl-	1268	1306	1999
000556-24-1	Methyl isovalerate	761	777	970
000491-02-1	Cyclohexanol, 5-methyl-2-(1-methylethyl)-, (1α,2α,5α)-	1155	1174	1512
000093-15-2	Methyleugenol	1368	1405	1890
000628-97-7	Hexadecanoic acid, ethyl ester	1975	1993	2246
000620-02-0	2-Furancarboxaldehyde, 5-methyl-	928	963	1451
000698-76-0	2*H*-Pyran-2-one, tetrahydro-6-propyl-	1235	1287	2024
000105-54-4	Butanoic acid, ethyl ester	787	804	1045
000103-38-8	Butanoic acid, 3-methyl-, phenylmethyl ester	1364	1396	1816
060784-31-8	2-Nonenal, (*Z*)-	1134	1159	1520
000105-68-0	1-Butanol, 3-methyl-, propanoate	952	968	1168
000106-27-4	Butanoic acid, 3-methylbutyl ester	1039	1055	1255
000103-45-7	Acetic acid, 2-phenylethyl ester	1223	1258	1712
000103-28-6	Propanoic acid, 2-methyl-, phenylmethyl ester	1268	1298	1702
000101-84-8	Diphenyl ether	1369	1404	1863
000106-33-2	Dodecanoic acid, ethyl ester	1576	1593	1827
000122-97-4	3-Phenylpropanol	1198	1233	1824
000112-44-7	Undecanal	1285	1306	1575
000536-59-4	1-Cyclohexene-1-methanol, 4-(1-methylethenyl)-	1274	1301	1791
000093-08-3	2-Naphthyl methyl ketone	1568	1620	2440
000122-69-0	Cinnamyl cinnamate	2347	2416	3127
000140-39-6	Acetic acid, 4-methylphenyl ester	1138	1170	1590
000104-21-2	Benzenemethanol, 4-methoxy-, acetate	1377	1421	2004
007452-79-1	Butanoic acid, 2-methyl-, ethyl ester	835	849	1028
000111-13-7	2-Octanone	969	991	1275
000513-86-0	Acetoin	*677*	714	1118
000710-04-3	2*H*-Pyran-2-one, 6-hexyltetrahydro-	1555	1607	2396
000110-45-2	1-Butanol, 3-methyl-, formate	774	792	1006
006728-26-3	2-Hexenal, (*E*)-	827	852	1187
000077-53-2	Cedrol	1583	1611	2072
000123-68-2	Hexanoic acid, 2-propenyl ester	1061	1079	1318

Table 1 continued

CAS	Chemical name	HP-1	HP5-MS	DB-225MS
002035-99-6	Octanoic acid, 3-methylbutyl ester	1428	1445	1641
000065-85-0	Benzoic acid	1155	1178	1314
000103-52-6	β-Phenylethyl butyrate	1408	1443	1896
000109-21-7	Butanoic acid, butyl ester	979	995	1199
000119-61-9	Benzophenone	1583	1635	2372
000821-55-6	2-Nonanone	1070	1091	1383
000109-15-9	Propanoic acid, 2-methyl-, octyl ester	1329	1344	1531
000108-64-5	Butanoic acid, 3-methyl-, ethyl ester	838	852	1038
000120-57-0	Piperonal	1286	1336	2043
000077-93-0	Triethyl citrate	1618	1664	2369
000623-17-6	2-Furanmethanol, acetate	964	995	1368
000104-57-4	Formic acid, phenylmethyl ester	1045	1078	1520
000124-07-2	Octanoic acid	1164	1179	1652
002244-16-8	D-Carvone	1212	1246	1701
000093-04-9	Naphthalene, 2-methoxy-	1415	1455	2051
031502-14-4	2-Nonen-1-ol, (*E*)-	1150	1168	1517
000111-13-7	2-Octanone	969	991	1274
000503-74-2	Butanoic acid, 3-methyl-	839	850	1286
000100-52-7	Benzaldehyde	929	961	1399
000928-95-0	2-Hexen-1-ol, (*E*)-	848	865	1202
000102-76-1	Triacetin	1309	1352	1957
000118-61-6	Benzoic acid, 2-hydroxy-, ethyl ester	1243	1273	1697
000112-06-1	Acetic acid, heptyl ester	1094	1111	1335
000093-04-9	Naphthalene, 2-methoxy-	1414	1454	2050
000140-27-2	Butanoic acid, 3-methyl-, 3-phenyl-2-propenyl ester	1648	1686	2226
000821-55-6	2-Nonanone	1070	1091	1383
000706-14-9	2(3*H*)-Furanone, 5-hexyldihydro-	1422	1471	2198
006728-26-3	2-Hexenal, (*E*)-	827	852	1187
007493-72-3	Allyl nonanoate	1358	1377	1593
007779-65-9	Isoamyl cinnamate	1705	1745	2304
000123-92-2	1-Butanol, 3-methyl-, acetate	859	876	1081
000112-30-1	1-Decanol	1254	1271	1577
000089-74-7	Ethanone, 1-(2,4-dimethylphenyl)-	1220	1253	1719
000823-22-3	2*H*-Pyran-2-one, tetrahydro-6-methyl-	1041	1095	1818
000103-37-7	Butanoic acid, phenylmethyl ester	1313	1347	1783
000544-12-7	3-Hexen-1-ol	838	855	1193
002497-18-9	2-Hexen-1-ol, acetate, (*E*)-	995	1015	1261
015356-70-4	Cyclohexanol, 5-methyl-2-(1-methylethyl)-, (1α,2α,5α)-	1155	1174	1512
000110-40-7	Decanedioic acid, diethyl ester	1751	1787	2293
000087-19-4	Benzoic acid, 2-hydroxy-, 2-methylpropyl ester	1444	1475	1920
000111-27-3	1-Hexanol	852	867	1179
000698-10-2	2(5*H*)-Furanone, 5-ethyl-3-hydroxy-4-methyl-	1158	1195	1974
000124-06-1	Tetradecanoic acid, ethyl ester	1775	1792	2036
000105-13-5	Benzenemethanol, 4-methoxy-	1244	1284	1994
000110-43-0	2-Heptanone	868	891	1166
000334-48-5	*n*-Decanoic acid	1354	1370	1885
000591-68-4	Pentanoic acid, butyl ester	1075	1093	1302
000121-32-4	Ethyl Vanillin	1407	1459	2315
000120-51-4	Benzyl Benzoate	1717	1770	2493

Table 1 continued

CAS	Chemical name	HP-1	HP5-MS	DB-225MS
000112-05-0	Nonanoic acid	1260	1274	1777
000119-36-8	Methyl salicylate	1166	1196	1594
000638-25-5	Pentyl octanoate	1466	1484	1714
003681-71-8	3-Hexen-1-ol, acetate, (*Z*)-	987	1006	1252
000100-86-7	Benzeneethanol, α,α-dimethyl-	1130	1158	1581
000110-43-0	2-Heptanone	868	891	1166
000110-27-0	Isopropyl myristate	1809	1824	2037
000103-48-0	Propanoic acid, 2-methyl-, 2-phenylethyl ester	1367	1396	1818
000593-08-8	2-Tridecanone	1474	1495	1818
000102-16-9	Benzeneacetic acid, phenylmethyl ester	1759	1815	2576
000112-17-4	Acetic acid, decyl ester	1391	1409	1617
000659-70-1	Butanoic acid, 3-methyl-, 3-methylbutyl ester	1090	1105	1292
000112-12-9	2-Undecanone	1272	1293	1573
000118-58-1	Benzoic acid, 2-hydroxy-, phenylmethyl ester	1823	1875	2633
004940-11-8	4*H*-Pyran-4-one, 2-ethyl-3-hydroxy-	1161	1197	1777
000591-12-8	2(3*H*)-Furanone, 5-methyl-	833	869	1325
000108-29-2	2(3*H*)-Furanone, dihydro-5-methyl-	905	953	1569
000104-93-8	Benzene, 1-methoxy-4-methyl-	997	1021	1333
000099-87-6	*p*-Cymene	1010	1025	1218
000106-30-9	Heptanoic acid, ethyl ester	1081	1127	1305
000106-65-0	Butanedioic acid, dimethyl ester	999	1032	1467
000124-06-1	Tetradecanoic acid, ethyl ester	1775	1793	2036
000123-92-2	1-Butanol, 3-methyl-, acetate	860	876	1080
000104-50-7	2(3*H*)-Furanone, 5-butyldihydro-	1210	1259	1949
003301-94-8	δ-Nonalactone	1343	1394	2148
010031-82-0	Benzaldehyde, 4-ethoxy-	1288	1333	1955
015356-74-8	2(4*H*)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-	1481	1537	2477
000151-10-0	Benzene, 1,3-dimethoxy-	1135	1168	1580
000123-29-5	Nonanoic acid, ethyl ester	1278	1295	1509
000111-11-5	Octanoic acid, methyl ester	1106	1125	1347
000706-14-9	2(3*H*)-Furanone, 5-hexyldihydro-	1422	1471	2197
000093-89-0	Benzoic acid, ethyl ester	1142	1172	1547
000127-41-3	α-Ionone	1403	1431	1875
000705-86-2	2*H*-Pyran-2-one, tetrahydro-6-pentyl-	1448	1499	2270
000109-25-1	Heptanoic acid, 3-methylbutyl ester	1330	1347	1876
000112-63-0	9,12-Octadecadienoic acid (*Z,Z*)-, methylester	2067	2094	2466
016409-46-4	Menthyl isovalerate	1500	1518	1744
001076-56-8	Benzene, 2-methoxy-4-methyl-1-(1-methylethyl)-	1213	1235	1503
004411-89-6	Benzeneacetaldehyde, α-ethylidene-	1233	1274	1858
007452-79-1	Butanoic acid, 2-methyl-, ethyl ester	836	849	1027
004536-23-6	Hexanoic acid, 2-methyl-	1027	1043	1484
000088-69-7	Phenol, 2-(1-methylethyl)-	1174	1199	1788
003452-97-9	1-Hexanol, 3,5,5-trimethyl-	1033	1047	1358
000104-53-0	Benzenepropanal	1122	1163	1693
000104-67-6	2(3*H*)-Furanone, 5-heptyldihydro-	1527	1576	2321
000695-06-7	2(3*H*)-Furanone, 5-ethyldihydro-	1005	1055	1708
000638-49-3	Formic acid, pentyl ester	811	826	1050
033467-73-1	3-Hexen-1-ol, formate, (*Z*)-	902	920	1177
000090-05-1	Phenol, 2-methoxy-	1058	1090	1577

Table 1 continued

CAS	Chemical name	HP-1	HP5-MS	DB-225MS
000122-84-9	2-Propanone, 1-(4-methoxyphenyl)-	1339	1386	2066
013360-65-1	Pyrazine, 3-ethyl-2,5-dimethyl-	1055	1078	1370
004748-78-1	Benzaldehyde, 4-ethyl-	1145	1179	1636
000087-44-5	Caryophyllene	1413	1426	1586
000103-50-4	Benzene, 1,1'-[oxybis(methylene)]bis-	1610	1654	2256
001731-84-6	Nonanoic acid, methyl ester	1205	1223	1452
000124-06-1	Tetradecanoic acid, ethyl ester	1775	1792	2036
003681-71-8	3-Hexen-1-ol, acetate, (*Z*)-	987	1006	1251
000539-82-2	Pentanoic acid, ethyl ester	837	852	1037
000108-21-4	Isopropyl acetate	*603*	*589*	844
000659-70-1	Butanoic acid, 3-methyl-, 3-methylbutyl ester	1090	1104	1292
006753-98-6	Humulene	1445	1460	1916
025152-84-5	2,4-Decadienal, (*E,E)*-	1287	1316	1776
003268-49-3	Methional	865	907	1368
001125-21-9	2,6,6-Trimethyl-2-cyclohexene-1,4-dione	1104	1144	1646
021834-92-4	5-Methyl-2-phenyl-2-hexenal	1455	1493	2065
019464-95-0	Oxiranecarboxylic acid, 3-methyl-3-phenyl-, ethyl ester, *cis*-	1489	1503	2007
002548-87-0	2-Octenal, (*E*)-	1032	1057	1414
000122-91-8	Benzenemethanol, 4-methoxy-, formate	1291	1334	1933
000124-19-6	Nonanal	1082	1104	1383
000104-67-6	2(3*H*)-Furanone, 5-heptyldihydro-	1528	1576	2321
000590-86-3	Butanal, 3-methyl-	870	900	897
000077-83-8	Oxiranecarboxylic acid, 3-methyl-3-phenyl-, ethyl ester	1489	1531	2142
014371-10-9	Cinnamaldehyde, (*E*)-	1227	1272	1922
000112-12-9	2-Undecanone	1272	1293	1573
000122-03-2	Benzaldehyde, 4-(1-methylethyl)-	1209	1242	1716
000107-75-5	Octanal, 7-hydroxy-3,7-dimethyl-	1254	1288	1864
002385-77-5	6-Octenal, 3,7-dimethyl-, (*R*)-	1130	1153	1460
000122-40-7	Cinnamaldehyde, α-pentyl-	1613	1651	2206
035044-68-9	2-Buten-1-one, 1-(2,6,6-trimethyl-1-cyclohexen-1-yl)-	1390	1418	1832
024720-09-0	2-Buten-1-one, 1-(2,6,6-trimethyl-2-cyclohexen-1-yl)-, (*E*)-	1370	1393	1789
004313-03-5	2,4-Heptadienal, (*E,E*)-	981	1010	1430
038462-22-5	Cyclohexanone, 2-(1-mercapto-1-methylethyl)-5-methyl-	1331	1367	1880
000079-69-6	α-Irone	1491	1519	1982
000689-67-8	5,9-Undecadien-2-one, 6,10-dimethyl-	1408	1434	1804
000112-45-8	10-Undecenal	1274	1299	1609
000105-43-1	Pentanoic acid, 3-methyl-	933	944	1297
000122-00-9	Ethanone, 1-(4-methylphenyl)-	1151	1186	1679
003796-70-1	5,9-Undecadien-2-one, 6,10-dimethyl-, (*E*)-	1426	1453	1831
000090-02-8	Benzaldehyde, 2-hydroxy-	1009	1044	1504
000106-72-9	5-Heptenal, 2,6-dimethyl-	1033	1054	1498
000112-54-9	Dodecanal	1387	1408	1713
002548-87-0	2-Octenal, (*E*)-	1021	1057	1441
006728-31-0	4-Heptenal, (*Z*)-	874	901	1194
001196-31-2	Cyclohexanone, 5-methyl-2-(1-methylethyl)-, (2*R-cis*)-	1130	1155	1521
000098-86-2	Acetophenone	1032	1067	1532
000066-25-1	Hexanal	778	803	1057
000110-62-3	Pentanal	*673*	704	948

Table 1 continued

CAS	Chemical name	HP-1	HP5-MS	DB-225MS
000103-95-7	3-(4-Isopropylphenyl)-2-methylpropionaldehyde	1426	1464	1959
000105-57-7	Ethane, 1,1-diethoxy-	721	729	1002
003391-86-4	1-Octen-3-ol	963	978	1278
003268-49-3	Methional	865	907	1368
000112-44-7	Undecanal	1285	1306	1575
000111-87-5	1-Octanol	1054	1069	1391
000111-27-3	1-Hexanol	852	867	1178
000102-19-2	Acetic acid, phenyl-, isopentyl ester	1470	1497	1950
000103-56-0	2-Propen-1-ol, 3-phenyl-, propanoate	1515	1555	2105
000141-14-0	6-Octen-1-ol, 3,7-dimethyl-, propanoate	1429	1448	1705
000105-86-2	2,6-Octadien-1-ol, 3,7-dimethyl-, formate, (*E*)-	1284	1306	1564
000106-29-6	Butanoic acid, 3,7-dimethyl-2,6-octadienyl ester, (*E*)-	1539	1563	1855
013532-18-8	3-(Methylthio)propanoic acid methyl ester	993	1024	1424
000103-73-1	Benzene, ethoxy-	969	993	1286
000110-27-0	Isopropyl myristate	1809	1824	2037
000124-07-2	Octanoic acid	1163	1179	1647
001188-02-9	2-Methylheptanoic acid	1128	1141	1564
000104-76-7	1-Hexanol, 2-ethyl-	1014	1028	1333
004437-51-8	3,4-Hexanedione	777	802	1083
001577-18-0	3-Hexenoic acid, (*E*)-	988	1003	1508
000079-09-4	Propanoic acid	*648*	706	1138
000111-14-8	Heptanoic acid	1070	1084	1545
016409-43-1	2*H*-Pyran, tetrahydro-4-methyl-2-(2-methyl-1-propenyl)-	1095	1111	1326
000078-83-1	1-Propanol, 2-methyl-	*564*	*593*	916
000079-31-2	Propanoic acid, 2-methyl-	754	765	1181
000106-25-2	2,6-Octadien-1-ol, 3,7-dimethyl-, (*Z*)-	1208	1228	1582
007212-44-4	1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-	1515	1534	1866
000111-87-5	1-Octanol	1054	1069	963
003681-71-8	3-Hexen-1-ol, acetate, (*Z*)-	964	1006	1391
000123-51-3	1-Butanol, 3-methyl-	722	734	1251
000100-51-6	Benzyl alcohol	1004	1034	1033
000123-66-0	Hexanoic acid, ethyl ester	981	999	1202
000143-13-5	Acetic acid, nonyl ester	1292	1309	1532
000150-84-5	6-Octen-1-ol, 3,7-dimethyl-, acetate	1333	1352	1584
000140-26-1	Butanoic acid, 3-methyl-, 2-phenylethyl ester	1461	1494	1931
000111-62-6	Ethyl Oleate	2144	2167	2475
000103-45-7	Acetic acid, 2-phenylethyl ester	1223	1257	1709
000106-22-9	Citronellol	1208	1227	1566
000111-12-6	2-Octynoic acid, methyl ester	1169	1202	1557
000105-53-3	Propanedioic acid, diethyl ester	1038	1070	1477
000105-66-8	Butanoic acid, propyl ester	881	899	1096
000141-16-2	Citronellyl butyrate	1508	1528	1784
000539-90-2	Butanoic acid, 2-methylpropyl ester	939	954	1145
024851-98-7	Cyclopentaneacetic acid, 3-oxo-2-pentyl-, methyl ester	1613	1657	2287
002198-61-0	Isopentyl hexanoate	1232	1249	1458
000105-85-1	6-Octen-1-ol, 3,7-dimethyl-, formate	1256	1275	1540
000106-22-9	Citronellol	1208	1227	1566
000078-70-6	1,6-Octadien-3-ol, 3,7-dimethyl-	1083	1099	1396
013532-18-8	3-(Methylthio)propanoic acid methyl ester	993	1024	1424

Table 1 continued

CAS	Chemical name	HP-1	HP5-MS	DB-225MS
000623-17-6	2-Furanmethanol, acetate	964	995	1366
000104-57-4	Formic acid, phenylmethyl ester	1045	1078	1518
005655-61-8	Bicyclo[2.2.1]heptan-2-ol, 1,7,7-trimethyl-, acetate, (1*S-endo*)-	1268	1288	1561
000103-59-3	Propanoic acid, 2-methyl-, 3-phenyl-2-propenyl ester	1547	1584	2105
000103-37-7	Butanoic acid, phenylmethyl ester	1313	1347	1781
000101-41-7	Benzeneacetic acid, methyl ester	1144	1178	1598
000140-88-5	2-Propenoic acid, ethyl ester	*649*	702	911
000105-85-1	6-Octen-1-ol, 3,7-dimethyl-, formate	1256	1275	1540
059020-85-8	Propanethioic acid, *S*-(2-furanylmethyl) ester	1219	1256	1689
000109-60-4	*n*-Propyl acetate	703	715	918
000093-92-5	Benzenemethanol,α-methyl-, acetate	1164	1194	1571
000125-12-2	Isobornyl acetate	1268	1290	1569
000109-19-3	Butanoic acid, 3-methyl-, butyl ester	1027	1045	1238
000123-69-3	Oxacycloheptadec-8-en-2-one, (8*Z*)	1900	1937	2459
020665-85-4	Propanoic acid, 2-methyl-, 4-formyl-2-methoxyphenyl ester	1613	1671	2470
000105-21-5	2(3*H*)-Furanone, dihydro-5-propyl-	1105	1154	1822
000107-92-6	Butanoic acid	779	794	1125
000765-70-8	1,2-Cyclopentanedione, 3-methyl-	994	1027	1547
023747-48-0	5*H*-5-Methyl-6,7-dihydrocyclopentapyrazine	1107	1140	1529
018138-04-0	Pyrazine, 2,3-diethyl-5-methyl-	1132	1155	1415
013678-59-6	Furan, 2-methyl-5-(methylthio)-	930	951	1215
015707-23-0	Pyrazine, 2-ethyl-3-methyl-	978	1003	1307
013925-00-3	Pyrazine, ethyl-	890	915	1228
001759-28-0	Thiazole, 5-ethenyl-4-methyl-	999	1026	1387
015707-23-0	Pyrazine, 2-ethyl-3-methyl-	978	1003	1308
024683-00-9	Pyrazine, 2-methoxy-3-(2-methylpropyl)-	1160	1181	1414
000693-95-8	4-Methylthiazole	793	817	1136
000137-00-8	5-Thiazoleethanol, 4-methyl-	1233	1277	2049
001759-28-0	Thiazole, 5-ethenyl-4-methyl-	999	1026	1387
000127-91-3	β-Pinene	968	978	1092
001438-91-1	Furan, 2-[(methylthio)methyl]-	972	1001	1352
000098-02-2	2-Furfurylthiol	883	911	1259
001124-11-4	Pyrazine, tetramethyl-	1063	1087	1381
014667-55-1	Pyrazine, trimethyl-	979	1004	1293
002882-20-4	Pyrazine, 2-methyl-3-(methylthio)-	1139	1169	1521
034413-35-9	5,6,7,8-Tetrahydroquinoxaline	1173	1210	1638
002884-13-1	Pyrazine, 2-methyl-6-(methylthio)-	1155	1187	1583
000350-03-8	Ethanone, 1-(3-pyridinyl)-	1074	1111	1705
001193-79-9	2-Acetyl-5-methylfuran	1005	1038	1507
022047-25-2	Acetylpyrazine	987	1022	1470
024295-03-2	2-Acetylthiazole	981	1019	1476
005910-89-4	Pyrazine, 2,3-dimethyl-	893	918	1240
000093-51-6	Creosol	1163	1193	1715
025680-58-4	2-Ethyl-3-methoxypyrazine	1032	1053	1301
024683-00-9	Pyrazine, 2-methoxy-3-(2-methylpropyl)-	1160	1181	1414
013925-07-0	Pyrazine, 2-ethyl-3,5-dimethyl-	1060	1084	1370
001122-62-9	Ethanone, 1-(2-pyridinyl)-	999	1033	1458
000093-16-3	Benzene, 1,2-dimethoxy-4-(1-propenyl)-	1418	1457	2050
000093-15-2	Methyleugenol	1367	1404	1887

Table 1 continued

CAS	Chemical name	HP-1	HP5-MS	DB-225MS
000499-75-2	Phenol, 2-methyl-5-(1-methylethyl)-	1265	1291	1866
000119-84-6	Hydrocoumarin	1327	1387	2197
005989-27-5	D-Limonene	1020	1029	1151
000097-53-0	Eugenol	1325	1359	1920
000513-86-0	Acetoin	*693*	714	1116
005932-68-3	*trans*-Isoeugenol	1413	1451	2092
034413-35-9	5,6,7,8-Tetrahydroquinoxaline	1171	1208	1636
000099-83-2	α-Phellandrene	995	1006	1128
000515-13-9	Cyclohexane, 1-ethenyl-1-methyl-2, 4-bis (1-methylethenyl)-, [1*S*-(1α,2β,4β)]-	1384	1426	1545
000586-62-9	Cyclohexene, 1-methyl-4-(1-methylethylidene)-	1077	1089	1229
000123-35-3	β-Myrcene	982	991	1107
000540-07-8	Hexanoic acid, pentyl ester	1269	1287	1506
000106-02-5	Oxacyclohexadecan-2-one	1805	1839	2591
000111-87-5	1-Octanol	1054	1070	1392
000078-59-1	Isophorone	1089	1122	1581
000470-82-6	Eucalyptol	1018	1032	1212
000060-12-8	Phenylethyl Alcohol	1082	1114	1660
007549-33-9	Anisyl propionate	1471	1514	2092
000110-38-3	Decanoic acid, ethyl ester	1377	1394	1586
000118-58-1	Benzoic acid, 2-hydroxy-, phenylmethyl ester	1825	1877	2634

The experimental conditions used for determination of the retention indices in Table 1 were: initial temperature 50 °C for 1 min, then increased at 6 °C min^{-1} to 280 °C, which was maintained for 2 min. The gas flow rate was 1.0 mL min^{-1}. Italics are used to indicate retention indices estimated by extrapolation

process. △RI for homologous compounds under different experimental conditions were also checked, and similar results were obtained.

Effect of Experimental Conditions

Although RI are regarded as being independent of operating conditions, except for stationary phase polarity [21], there are reports of RI varying with column geometry, carrier gas flow-rate, initial temperature, and heating rate [24]. In fact, despite substantial improvements in instrumentation, acquisition of reliable retention indices may still be difficult. Consequently, misidentification may occur when retention indices are used as reference values, especially retention indices from different published studies. To overcome this problem, some authors have proposed use of techniques for conversion of retention indices obtained under different conditions to furnish standardized RI for interlaboratory comparison [24]. However, the problem of RI standardization has not yet been resolved, although many theories have been proposed on the basis of thermodynamics or quantitative structure–retention relationships.

In this work, taking into account that different experimental conditions may be required when different samples are analyzed, some experimental conditions which are often varied, gas flow rate, heating rate, and initial temperature, were considered during development of the retention index database. The HP-5MS capillary column was used for this experiment. RI of some representative aroma compounds under different experimental conditions are listed in Tables 2 and 3.

From Table 2 it is apparent the maximum difference between RI values for heating rates of 4, 6, 8, and 10 °C min^{-1} is 10 retention index units for most of the compounds. This indicates the retention indices are very stable when different heating rates are used, which ensures identification by use of RI matching is reliable. However, it is worth noting there are 22 compounds, including 2-naphthyl methyl ketone, benzophenone, humulene, and oxacyclohexadecan-2-one, for which △RI is >10 retention index units. All these 22 molecules contain a benzene ring or a macrocyclic substructure, but the reason for the difference is unclear. Details about molecular structure and the corresponding retention indices can be found in the supporting information (supplementary Table 5).

For most of the compounds, values of RI increase with the increasing heating rate. In contrast, the values of RI for acids decrease with increasing heating rate. For example,

Table 2 Retention indices on HP-5MS, obtained by use of different heating rates

CAS	Chemical name	4°/min	6°/min	8°/min	10°/min
000102-20-5	Benzeneacetic acid, 2-phenylethyl ester	1914	1922	1926	1931
000112-14-1	Acetic acid, octyl ester	1210	1210	1210	1210
000093-18-5	Naphthalene, 2-ethoxy-	1522	1528	1532	1536
000102-13-6	Benzeneacetic acid, 2-methylpropyl ester	1390	1392	1393	1395
001504-74-1	2-Propenal, 3-(2-methoxyphenyl)-	1528	1533	1535	1537
000106-32-1	Octanoic acid, ethyl ester	1196	1196	1196	1196
000097-62-1	Propanoic acid, 2-methyl-, ethyl ester	757	757	758	758
000695-06-7	2(3*H*)-Furanone, 5-ethyldihydro-	1052	1054	1055	1057
000112-31-2	Decanal	1204	1205	1205	1206
000488-10-8	2-Cyclopenten-1-one, 3-methyl-2-(2-pentenyl)-, (*Z*)-	1398	1401	1403	1406
000110-19-0	Isobutyl acetate	773	773	774	774
002721-22-4	2*H*-Pyran-2-one, tetrahydro-6-nonyl-	1921	1926	1929	1932
000123-69-3	Oxacycloheptadec-8-en-2-one, (8*Z*)	1928	1938	1945	1951
000556-82-1	2-Buten-1-ol, 3-methyl-	775	776	776	777
000106-29-6	Butanoic acid, 3,7-dimethyl-2,6-octadienyl ester, (*E*)-	1560	1560	1560	1561
005471-51-2	2-Butanone, 4-(4-hydroxyphenyl)-	1550	1553	1553	1553
000104-54-1	2-Propen-1-ol, 3-phenyl-	1304	1306	1308	1310
000556-24-1	Methyl isovalerate	776	777	777	777
000491-02-1	Cyclohexanol, 5-methyl-2-(1-methylethyl)-, (1α,2α,5α)-	1171	1174	1175	1177
000093-15-2	Methyleugenol	1404	1405	1406	1406
000628-97-7	Hexadecanoic acid, ethyl ester	1992	1993	1993	1993
000620-02-0	2-Furancarboxaldehyde, 5-methyl-	961	963	964	965
000698-76-0	2*H*-Pyran-2-one, tetrahydro-6-propyl-	1283	1287	1288	1291
000105-54-4	Butanoic acid, ethyl ester	804	804	804	804
000103-38-8	Butanoic acid, 3-methyl-, phenylmethyl ester	1394	1396	1397	1399
060784-31-8	2-Nonenal, (*Z*)-	1158	1159	1160	1161
000105-68-0	1-Butanol, 3-methyl-, propanoate	968	968	968	969
000106-27-4	Butanoic acid, 3-methylbutyl ester	1055	1055	1055	1055
000103-45-7	Acetic acid, 2-phenylethyl ester	1256	1258	1259	1260
000103-28-6	Propanoic acid, 2-methyl-, phenylmethyl ester	1297	1298	1299	1301
000101-84-8	Diphenyl ether	1399	1404	1408	1411
000106-33-2	Dodecanoic acid, ethyl ester	1593	1593	1586	1593
000122-97-4	3-Phenylpropanol	1230	1233	1234	1236
000112-44-7	Undecanal	1305	1306	1306	1307
000536-59-4	1-Cyclohexene-1-methanol, 4-(1-methylethenyl)-	1298	1301	1302	1304
000093-08-3	2-Naphthyl methyl ketone	1612	1620	1626	1631
000122-69-0	Cinnamyl cinnamate	2407	2416	2422	2428
000140-39-6	Acetic acid, 4-methylphenyl ester	1168	1170	1171	1172
000104-21-2	Benzenemethanol, 4-methoxy-, acetate	1419	1421	1422	1423
007452-79-1	Butanoic acid, 2-methyl-, ethyl ester	847	849	849	850
000111-13-7	2-Octanone	990	991	991	991
000513-86-0	Acetoin	713	714	714	713
000710-04-3	2*H*-Pyran-2-one, 6-hexyltetrahydro-	1603	1607	1609	1612
000110-45-2	1-Butanol, 3-methyl-, formate	792	792	792	792
006728-26-3	2-Hexenal, (*E*)-	850	852	853	854
000077-53-2	Cedrol	1602	1611	1617	1623
000123-68-2	Hexanoic acid, 2-propenyl ester	1081	1079	1080	1081
002035-99-6	Octanoic acid, 3-methylbutyl ester	1445	1445	1445	1446

Table 2 continued

CAS	Chemical name	4°/min	6°/min	8°/min	10°/min
000065-85-0	Benzoic acid	1179	1178	1177	1177
000103-52-6	β-Phenylethyl butyrate	1440	1443	1445	1446
000109-21-7	Butanoic acid, butyl ester	995	995	995	995
000119-61-9	Benzophenone	1627	1635	1639	1644
000821-55-6	2-Nonanone	1091	1091	1091	1092
000109-15-9	Propanoic acid, 2-methyl-, octyl ester	1344	1344	1344	1344
000108-64-5	Butanoic acid, 3-methyl-, ethyl ester	851	852	852	853
000120-57-0	Piperonal	1331	1336	1339	1342
000077-93-0	Triethyl citrate	1664	1664	1664	1664
000623-17-6	2-Furanmethanol, acetate	995	995	995	995
000104-57-4	Formic acid, phenylmethyl ester	1076	1078	1079	1080
000124-07-2	Octanoic acid	1181	1179	1177	1176
002244-16-8	D-Carvone	1243	1246	1248	1250
000093-04-9	Naphthalene, 2-methoxy-	1448	1455	1459	1463
031502-14-4	2-Nonen-1-ol, (*E*)-	1168	1168	1168	1169
000111-13-7	2-Octanone	990	991	991	991
000503-74-2	Butanoic acid, 3-methyl-	850	850	849	850
000928-95-0	2-Hexen-1-ol, (*E*)-	864	865	865	866
000102-76-1	Triacetin	1354	1352	1350	1350
000118-61-6	Benzoic acid, 2-hydroxy-, ethyl ester	1270	1273	1275	1277
000112-06-1	Acetic acid, heptyl ester	1112	1111	1111	1111
000093-04-9	Naphthalene, 2-methoxy-	1448	1454	1458	1462
000140-27-2	Butanoic acid, 3-methyl-, 3-phenyl-2-propenyl ester	1683	1686	1687	1690
000821-55-6	2-Nonanone	1091	1091	1091	1092
000706-14-9	2(3*H*)-Furanone, 5-hexyldihydro-	1468	1471	1473	1475
006728-26-3	2-Hexenal, (*E*)-	850	852	853	854
007493-72-3	Allyl nonanoate	1377	1377	1377	1377
007779-65-9	Isoamyl cinnamate	1741	1745	1747	1751
000123-92-2	1-Butanol, 3-methyl-, acetate	875	876	876	877
000112-30-1	1-Decanol	1270	1271	1270	1271
000089-74-7	Ethanone, 1-(2,4-dimethylphenyl)-	1250	1253	1255	1257
000823-22-3	2*H*-Pyran-2-one, tetrahydro-6-methyl-	1092	1095	1096	1098
000103-37-7	Butanoic acid, phenylmethyl ester	1345	1347	1348	1350
000544-12-7	3-Hexen-1-ol	853	855	855	856
002497-18-9	2-Hexen-1-ol, acetate, (*E*)-	1015	1015	1015	1015
015356-70-4	Cyclohexanol, 5-methyl-2-(1-methylethyl)-, (1α,2β,5α)-	1171	1174	1175	1177
000110-40-7	Decanedioic acid, diethyl ester	1787	1787	1786	1787
000087-19-4	Benzoic acid, 2-hydroxy-, 2-methylpropyl ester	1471	1475	1477	1480
000111-27-3	1-Hexanol	866	867	867	868
000698-10-2	2(5*H*)-Furanone, 5-ethyl-3-hydroxy-4-methyl-	1194	1195	1195	1197
000124-06-1	Tetradecanoic acid, ethyl ester	1792	1792	1792	1792
000105-13-5	Benzenemethanol, 4-methoxy-	1281	1284	1285	1287
000110-43-0	2-Heptanone	890	891	891	891
000334-48-5	*n*-Decanoic acid	1372	1370	1368	1367
000591-68-4	Pentanoic acid, butyl ester	1093	1093	1092	1092
000121-32-4	Ethyl Vanillin	1455	1459	1461	1463
000120-51-4	Benzyl Benzoate	1764	1770	1774	1779
000112-05-0	Nonanoic acid	1277	1274	1272	1272
000119-36-8	Methyl salicylate	1193	1196	1198	1200

Table 2 continued

CAS	Chemical name	4°/min	6°/min	8°/min	10°/min
000638-25-5	Pentyl octanoate	1483	1484	1483	1484
003681-71-8	3-Hexen-1-ol, acetate, (*Z*)-	1006	1006	1006	1006
000100-86-7	Benzeneethanol, α,α-dimethyl-	1155	1158	1159	1161
000110-43-0	2-Heptanone	890	891	891	891
000110-27-0	Isopropyl myristate	1824	1824	1823	1824
000103-48-0	Propanoic acid, 2-methyl-, 2-phenylethyl ester	1394	1396	1398	1399
000593-08-8	2-Tridecanone	1494	1495	1495	1496
000102-16-9	Benzeneacetic acid, phenylmethyl ester	1809	1815	1819	1824
000112-17-4	Acetic acid, decyl ester	1409	1409	1409	1409
000659-70-1	Butanoic acid, 3-methyl-, 3-methylbutyl ester	1105	1105	1104	1105
000112-12-9	2-Undecanone	1292	1293	1293	1293
000118-58-1	Benzoic acid, 2-hydroxy-, phenylmethyl ester	1868	1875	1880	1885
004940-11-8	4*H*-Pyran-4-one, 2-ethyl-3-hydroxy-	1195	1197	1199	1201
000591-12-8	2(3*H*)-Furanone, 5-methyl-	867	869	869	871
000108-29-2	2(3*H*)-Furanone, dihydro-5-methyl-	950	953	953	955
000104-93-8	Benzene, 1-methoxy-4-methyl-	1019	1021	1022	1023
000099-87-6	*p*-Cymene	1023	1025	1027	1028
000106-30-9	Heptanoic acid, ethyl ester	1098	1098	1097	1097
000106-65-0	Butanedioic acid, dimethyl ester	1032	1032	1032	1032
000124-06-1	Tetradecanoic acid, ethyl ester	1792	1793	1792	1793
000123-92-2	1-Butanol, 3-methyl-, acetate	875	876	876	877
000104-50-7	2(3*H*)-Furanone, 5-butyldihydro-	1257	1259	1261	1262
003301-94-8	δ-Nonalactone	1391	1394	1396	1398
010031-82-0	Benzaldehyde, 4-ethoxy-	1329	1333	1334	1337
015356-74-8	2(4*H*)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-	1529	1537	1483	1547
000151-10-0	Benzene, 1,3-dimethoxy-	1166	1168	1169	1170
000123-29-5	Nonanoic acid, ethyl ester	1295	1295	1295	1295
000111-11-5	Octanoic acid, methyl ester	1124	1125	1123	1124
000706-14-9	2(3*H*)-Furanone, 5-hexyldihydro-	1468	1471	1473	1475
000093-89-0	Benzoic acid, ethyl ester	1170	1172	1173	1174
000705-86-2	2*H*-Pyran-2-one, tetrahydro-6-pentyl-	1495	1499	1501	1504
000109-25-1	Heptanoic acid, 3-methylbutyl ester	1347	1347	1347	1350
000112-63-0	9,12-Octadecadienoic acid (*Z,Z*)-, methylester	2092	2094	2094	2097
016409-46-4	Menthyl isovalerate	1515	1518	1519	1521
001076-56-8	Benzene, 2-methoxy-4-methyl-1-(1-methylethyl)-	1234	1235	1236	1237
004411-89-6	Benzeneacetaldehyde, α-ethylidene-	1271	1274	1276	1278
007452-79-1	Butanoic acid, 2-methyl-, ethyl ester	848	849	849	850
004536-23-6	Hexanoic acid, 2-methyl-	1040	1043	1042	1041
000088-69-7	Phenol, 2-(1-methylethyl)-	1199	1199	1199	1199
003452-97-9	1-Hexanol, 3,5,5-trimethyl-	1045	1047	1047	1048
000104-53-0	Benzenepropanal	1161	1163	1165	1167
000104-67-6	2(3*H*)-Furanone, 5-heptyldihydro-	1572	1576	1577	1580
000695-06-7	2(3*H*)-Furanone, 5-ethyldihydro-	1052	1055	1056	1057
000638-49-3	Formic acid, pentyl ester	825	826	827	827
033467-73-1	3-Hexen-1-ol, formate, (*Z*)-	919	920	920	921
000090-05-1	Phenol, 2-methoxy-	1088	1090	1091	1092
000122-84-9	2-Propanone, 1-(4-methoxyphenyl)-	1383	1386	1387	1390
013360-65-1	Pyrazine, 3-ethyl-2,5-dimethyl-	1076	1078	1079	1080

Table 2 continued

CAS	Chemical name	4°/min	6°/min	8°/min	10°/min
004748-78-1	Benzaldehyde, 4-ethyl-	1176	1179	1181	1184
000087-44-5	Caryophyllene	1420	1426	1430	1434
000103-50-4	Benzene, 1,1'-[oxybis(methylene)]bis-	1649	1654	1658	1662
001731-84-6	Nonanoic acid, methyl ester	1223	1223	1223	1223
000124-06-1	Tetradecanoic acid, ethyl ester	1792	1792	1792	1793
003681-71-8	3-Hexen-1-ol, acetate, (*Z*)-	1006	1006	1006	1006
000539-82-2	Pentanoic acid, ethyl ester	850	852	852	853
000659-70-1	Butanoic acid, 3-methyl-, 3-methylbutyl ester	1105	1104	1107	1105
006753-98-6	Humulene	1454	1460	1464	1468
025152-84-5	2,4-Decadienal, (*E,E*)-	1314	1316	1317	1318
003268-49-3	Methional	906	907	907	908
021834-92-4	5-Methyl-2-phenyl-2-hexenal	1489	1493	1495	1497
002548-87-0	2-Octenal, (*E*)-	1056	1057	1058	1059
000122-91-8	Benzenemethanol, 4-methoxy-, formate	1331	1334	1335	1337
000124-19-6	Nonanal	1103	1104	1104	1104
000122-40-7	Cinnamaldehyde, α-pentyl-	1647	1651	1654	1657
035044-68-9	2-Buten-1-one, 1-(2,6,6-trimethyl-1-cyclohexen-1-yl)-	1414	1418	1420	1423
024720-09-0	2-Buten-1-one, 1-(2,6,6-trimethyl-2-cyclohexen-1-yl)-, (*E*)-	1392	1393	1398	1400
004313-03-5	2,4-Heptadienal, (*E,E*)-	1009	1010	1011	1011
038462-22-5	Cyclohexanone, 2-(1-mercapto-1-methylethyl)-5-methyl-	1361	1367	1371	1375
000079-69-6	α-Irone	1537	1519	1545	1548
000689-67-8	5,9-Undecadien-2-one, 6,10-dimethyl-	1433	1434	1435	1436
002548-87-0	2-Octenal, (*E*)-	1056	1057	1058	1059
006728-31-0	4-Heptenal, (*Z*)-	900	901	901	901
001196-31-2	Cyclohexanone, 5-methyl-2-(1-methylethyl)-, (2*R-cis*)-	1152	1155	1157	1159
000098-86-2	Acetophenone	1064	1067	1068	1070
000066-25-1	Hexanal	803	803	803	803
000110-62-3	Pentanal	704	704	705	704
000105-57-7	Ethane, 1,1-diethoxy-	728	729	730	729
003391-86-4	1-Octen-3-ol	978	978	979	979
003268-49-3	Methional	906	907	907	908
000112-44-7	Undecanal	1306	1306	1307	1307
000111-87-5	1-Octanol	1069	1069	1069	1070
000111-27-3	1-Hexanol	866	867	867	868
013532-18-8	3-(Methylthio)propanoic acid methyl ester	1023	1024	1025	1026
000103-73-1	Benzene, ethoxy-	992	993	994	995
000110-27-0	Isopropyl myristate	1823	1824	1823	1824
000124-07-2	Octanoic acid	1180	1179	1176	1175
001188-02-9	2-Methylheptanoic acid	1142	1141	1139	1138
000104-76-7	1-Hexanol, 2-ethyl-	1027	1028	1028	1028
004437-51-8	3,4-Hexanedione	802	802	802	802
001577-18-0	3-Hexenoic acid, (*E*)-	1006	1003	1001	1000
000079-09-4	Propanoic acid	706	706	706	704
000111-14-8	Heptanoic acid	1086	1084	1082	1081
016409-43-1	2*H*-Pyran, tetrahydro-4-methyl-2-(2-methyl-1-propenyl)-	1110	1111	1112	1113
000079-31-2	Propanoic acid, 2-methyl-	764	765	764	764
000106-25-2	2,6-Octadien-1-ol, 3,7-dimethyl-, (*Z*)-	1227	1228	1229	1230
000111-87-5	1-Octanol	1069	1069	1069	1070
003681-71-8	3-Hexen-1-ol, acetate, (*Z*)-	1006	1006	1006	1006

Table 2 continued

CAS	Chemical name	4°/min	6°/min	8°/min	10°/min
000123-51-3	1-Butanol, 3-methyl-	733	734	734	734
000100-51-6	Benzyl alcohol	1032	1034	1034	1036
000123-66-0	Hexanoic acid, ethyl ester	999	999	998	999
000143-13-5	Acetic acid, nonyl ester	1309	1309	1309	1309
000150-84-5	6-Octen-1-ol, 3,7-dimethyl-, acetate	1352	1352	1352	1352
000140-26-1	Butanoic acid, 3-methyl-, 2-phenylethyl ester	1491	1494	1495	1497
000111-62-6	Ethyl Oleate	2165	2167	2167	2168
000103-45-7	Acetic acid, 2-phenylethyl ester	1256	1257	1258	1260
000106-22-9	Citronellol	1227	1227	1227	1227
000111-12-6	2-Octynoic acid, methyl ester	1202	1202	1201	1202
000105-53-3	Propanedioic acid, diethyl ester	1070	1070	1069	1070
000105-66-8	Butanoic acid, propyl ester	899	899	899	899
000141-16-2	Citronellyl butyrate	1527	1528	1528	1528
000539-90-2	Butanoic acid, 2-methylpropyl ester	953	954	954	955
024851-98-7	Cyclopentaneacetic acid, 3-oxo-2-pentyl-, methyl ester	1654	1657	1658	1660
002198-61-0	Isopentyl hexanoate	1249	1249	1249	1246
000078-70-6	1,6-Octadien-3-ol, 3,7-dimethyl-	1098	1099	1099	1100
013532-18-8	3-(Methylthio)propanoic acid methyl ester	1023	1024	1025	1026
000623-17-6	2-Furanmethanol, acetate	995	995	994	995
000104-57-4	Formic acid, phenylmethyl ester	1076	1078	1079	1080
005655-61-8	Bicyclo[2.2.1]heptan-2-ol, 1,7,7-trimethyl-, acetate, (1*S*-*endo*)-	1285	1288	1290	1292
000103-59-3	Propanoic acid, 2-methyl-, 3-phenyl-2-propenyl ester	1582	1584	1586	1587
000103-37-7	Butanoic acid, phenylmethyl ester	1345	1347	1348	1350
000101-41-7	Benzeneacetic acid, methyl ester	1177	1178	1179	1180
000140-88-5	2-Propenoic acid, ethyl ester	702	702	702	700
000105-85-1	6-Octen-1-ol, 3,7-dimethyl-, formate	1274	1275	1275	1276
059020-85-8	Propanethioic acid, *S*-(2-furanylmethyl) ester	1253	1256	1257	1257
000109-60-4	*n*-Propyl acetate	715	715	716	715
000093-92-5	Benzenemethanol, α-methyl-, acetate	1193	1194	1195	1196
000125-12-2	Isobornyl acetate	1286	1290	1292	1294
000109-19-3	Butanoic acid, 3-methyl-, butyl ester	1045	1045	1045	1046
000123-69-3	Oxacycloheptadec-8-en-2-one, (8*Z*)	1927	1937	1944	1950
020665-85-4	Propanoic acid, 2-methyl-, 4-formyl-2-methoxyphenyl ester	1667	1671	1672	1675
000105-21-5	2(3*H*)-Furanone, dihydro-5-propyl-	1151	1154	1155	1156
000107-92-6	Butanoic acid	795	794	794	794
000765-70-8	1,2-Cyclopentanedione, 3-methyl-	1025	1027	1028	1029
023747-48-0	5*H*-5-Methyl-6,7-dihydrocyclopentapyrazine	1137	1140	1142	1144
018138-04-0	Pyrazine, 2,3-diethyl-5-methyl-	1154	1155	1156	1157
013678-59-6	Furan, 2-methyl-5-(methylthio)-	909	910	911	912
015707-23-0	Pyrazine, 2-ethyl-3-methyl-	1002	1003	1004	1005
013925-00-3	Pyrazine, ethyl-	913	915	915	916
001759-28-0	Thiazole, 5-ethenyl-4-methyl-	1024	1026	1027	1029
015707-23-0	Pyrazine, 2-ethyl-3-methyl-	1002	1003	1004	1005
024683-00-9	Pyrazine, 2-methoxy-3-(2-methylpropyl)-	1179	1181	1181	1182
000693-95-8	4-Methylthiazole	815	817	817	818
000137-00-8	5-Thiazoleethanol, 4-methyl-	1274	1277	1278	1280
001759-28-0	Thiazole, 5-ethenyl-4-methyl-	1024	1026	1027	1029
000127-91-3	β-Pinene	975	978	980	981
001438-91-1	Furan, 2-[(methylthio)methyl]-	999	1001	1002	1003

Table 2 continued

CAS	Chemical name	4°/min	6°/min	8°/min	10°/min
000098-02-2	2-Furfurylthiol	910	911	912	913
001124-11-4	Pyrazine, tetramethyl-	1085	1087	1088	1089
014667-55-1	Pyrazine, trimethyl-	1002	1004	1004	1005
034413-35-9	5,6,7,8-Tetrahydroquinoxaline	1205	1210	1212	1215
002884-13-1	Pyrazine, 2-methyl-6-(methylthio)-	1184	1187	1190	1192
000350-03-8	Ethanone, 1-(3-pyridinyl)-	1109	1111	1112	1115
001193-79-9	2-Acetyl-5-methylfuran	1036	1038	1039	1040
022047-25-2	Acetylpyrazine	1020	1022	1023	1024
024295-03-2	2-Acetylthiazole	1017	1019	1020	1021
005910-89-4	Pyrazine, 2,3-dimethyl-	916	918	919	920
000093-51-6	Creosol	1191	1193	1194	1196
025680-58-4	2-Ethyl-3-methoxypyrazine	1052	1053	1054	1055
024683-00-9	Pyrazine, 2-methoxy-3-(2-methylpropyl)-	1179	1181	1181	1182
013925-07-0	Pyrazine, 2-ethyl-3,5-dimethyl-	1082	1084	1084	1086
001122-62-9	Ethanone, 1-(2-pyridinyl)-	1031	1033	1034	1036
000093-15-2	Methyleugenol	1403	1404	1405	1406
000119-84-6	Hydrocoumarin	1381	1387	1390	1394
005989-27-5	D-Limonene	1027	1029	1031	1032
000097-53-0	Eugenol	1356	1359	1361	1363
000513-86-0	Acetoin	713	714	714	713
005932-68-3	*trans*-Isoeugenol	1448	1451	1453	1456
034413-35-9	5,6,7,8-Tetrahydroquinoxaline	1204	1208	1211	1214
000099-83-2	α-Phellandrene	1004	1006	1007	1008
000515-13-9	Cyclohexane, 1-ethenyl-1-methyl-2,4-bis(1-methylethenyl)-, [1*S*-(1α,2β,4β)]-	1392	1426	1398	1401
000586-62-9	Cyclohexene, 1-methyl-4-(1-methylethylidene)-	1087	1089	1090	1092
000540-07-8	Hexanoic acid, pentyl ester	1287	1287	1287	1287
000106-02-5	Oxacyclohexadecan-2-one	1830	1839	1845	1852
000111-87-5	1-Octanol	1069	1070	1069	1070
000078-59-1	Isophorone	1118	1122	1123	1125
000470-82-6	Eucalyptol	1030	1032	1034	1036
000060-12-8	Phenylethyl Alcohol	1112	1114	1116	1117
007549-33-9	Anisyl propionate	1512	1514	1515	1517
000110-38-3	Decanoic acid, ethyl ester	1394	1394	1394	1394
000118-58-1	Benzoic acid, 2-hydroxy-, phenylmethyl ester	1869	1877	1881	1886

the values of RI for octanoic acid are 1,180, 1,179, 1,176, and 1,175 for heating rates of 4, 6, 8, and 10 °C min^{-1}, respectively. These observations provide a chance for further data mining of retention indices.

Table 3 gives RI values for 51 representative compounds measured with initial temperatures of 50, 60, 70, 80, and 90 °C. It is apparent that changes of the retention indices are very small: the difference is below 10 retention index units for all the compounds. For most of the compounds, RI values increase with increasing initial temperature. However, the study of Boswell shows that the isothermal retention index of anthracene is highly dependent on the temperature at which it is measured. Use of linear temperature-programmed retention indices assumes that a given compound always elutes at the same position between the two bracketing *n*-alkanes, but this is clearly not the case when experimental conditions are changed. So, to investigate the effect of initial temperature on elution order, the change of elution order for different initial temperatures was studied on the basis of a simple ranking method. Fifty-one samples and *n*-alkanes were ranked by retention time; details can be found in the supporting information (supplementary Table 6). Two kinds of change in elution order were observed. The first is position changes between the two bracketing *n*-alkanes, for example, the positions of phenetole and ethyl caproate changed between C_9 and C_{10}. The second is that a given compound

Table 3 Retention indices on HP-5MS, obtained by use of different initial temperatures

CAS No.	Chemical Name	50/°C	60/°C	70/°C	80/°C	90/°C
04313-03-5	2,4-Heptadienal, (*E,E*)-	1010	1010	1011	1012	1013
038462-22-5	Cyclohexanone, 2-(1-mercapto-1-methylethyl)-5-methyl-	1367	1367	1367	1368	1371
000079-69-6	α-Irone	1541	1541	1542	1543	1545
000689-67-8	5,9-Undecadien-2-one, 6,10-dimethyl-	1434	1434	1434	1434	1437
000112-45-8	10-Undecenal	1299	1299	1299	1300	1302
000105-43-1	Pentanoic acid, 3-methyl-	944	943	937	933	928
000122-00-9	Ethanone, 1-(4-methylphenyl)-	1186	1186	1186	1188	1191
000090-02-8	Benzaldehyde, 2-hydroxy-	1044	1044	1045	1047	1050
000106-72-9	5-Heptenal, 2,6-dimethyl-	1054	1054	1053	1054	1055
000112-54-9	Dodecanal	1408	1408	1408	1408	1411
002548-87-0	2-Octenal, (*E*)-	1057	1057	1057	1057	1058
006728-31-0	4-Heptenal, (Z)-	901	900	901	903	904
001196-31-2	Cyclohexanone, 5-methyl-2-(1-methylethyl)-, (2*R-cis*)-	1156	1156	1156	1157	1160
000098-86-2	Acetophenone	1067	1067	1068	1069	1072
000066-25-1	Hexanal	803	802	803	806	807
000103-95-7	3-(4-Isopropylphenyl)-2-methylpropionaldehyde	1464	1464	1464	1465	1467
003391-86-4	1-Octen-3-ol	978	978	977	977	977
003268-49-3	Methional	907	906	907	909	911
000112-44-7	Undecanal	1306	1307	1307	1307	1309
000111-87-5	1-Octanol	1069	1069	1068	1068	1068
000111-27-3	1-Hexanol	867	865	863	863	864
000102-19-2	Acetic acid, phenyl-, isopentyl ester	1497	1501	1500	1501	1504
000103-56-0	2-Propen-1-ol, 3-phenyl-, propanoate	1555	1556	1556	1557	1559
000141-14-0	6-Octen-1-ol, 3,7-dimethyl-, propanoate	1448	1449	1449	1449	1451
000105-86-2	2,6-Octadien-1-ol, 3,7-dimethyl-, formate, (*E*)-	1306	1307	1306	1307	1309
013532-18-8	3-(Methylthio)propanoic acid methyl ester	1024	1024	1024	1025	1025
000103-73-1	Benzene, ethoxy-	993	993	994	997	999
000110-27-0	Isopropyl myristate	1824	1825	1824	1825	1828
000124-07-2	Octanoic acid	1179	1179	1177	1177	1173
001188-02-9	2-Methylheptanoic acid	1141	1140	1137	1136	1132
000104-76-7	1-Hexanol, 2-ethyl-	1028	1028	1027	1026	1027
001577-18-0	3-Hexenoic acid, (*E*)-	1003	1003	998	992	987
000111-14-8	Heptanoic acid	1084	1084	1081	1077	1073
016409-43-1	2*H*-Pyran, tetrahydro-4-methyl-2-(2-methyl-1-propenyl)-	1111	1111	1112	1112	1114
000106-25-2	2,6-Octadien-1-ol, 3,7-dimethyl-, (*Z*)-	1228	1228	1229	1229	1231
007212-44-4	1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-	1534	1534	1534	1535	1537
000111-87-5	1-Octanol	1069	1069	1068	1068	1068
003681-71-8	3-Hexen-1-ol, acetate, (*Z*)-	1006	1006	1006	1006	1006
000100-51-6	Benzyl alcohol	1034	1034	1034	1034	1036
000123-66-0	Hexanoic acid, ethyl ester	999	999	998	998	999
000143-13-5	Acetic acid, nonyl ester	1309	1310	1309	1309	1311
000150-84-5	6-Octen-1-ol, 3,7-dimethyl-, acetate	1352	1353	1352	1352	1354
000140-26-1	Butanoic acid, 3-methyl-, 2-phenylethyl ester	1494	1494	1494	1494	1496
000111-62-6	Ethyl Oleate	2167	2166	2166	2167	2170
000103-45-7	Acetic acid, 2-phenylethyl ester	1257	1258	1258	1258	1260
000106-22-9	Citronellol	1227	1227	1227	1227	1228
000111-12-6	2-Octynoic acid, methyl ester	1202	1202	1202	1202	1204
000105-53-3	Propanedioic acid, diethyl ester	1070	1070	1068	1067	1066
000105-66-8	Butanoic acid, propyl ester	899	898	897	897	898

Table 3 continued

CAS No.	Chemical Name	50/°C	60/°C	70/°C	80/°C	90/°C
000141-16-2	Citronellyl butyrate	1528	1528	1528	1528	1530
024851-98-7	Cyclopentaneacetic acid, 3-oxo-2-pentyl-, methyl ester	1657	1657	1656	1657	1659

The experimental conditions used for determination of the retention indices in Table 3 were: initial temperature 50, 60, 70, 80, or 90 °C for 2 min, then increased at 6 °C min^{-1} to 280 °C which was maintained for 2 min. The gas flow rate was 1 mL min^{-1}

elutes in a different *n*-alkanes interval; for example, 3-hexenoic acid elutes between C_{10} and C_{11} when the initial temperature is 50 or 60 °C but between C_9 or C_{10} when the initial temperature is 70, 80, or 90 °C. Similar results are obtained for undecylenic aldehyde and isoamyl phenylacetate. This is because of a large difference between the enthalpy of transfer of anthracene and the bracketing *n*-alkanes.

Finally, the effect on RI of carrier gas flow rate was also investigated. The results showed that absolute differences between RI values were between 0 and 8 retention index units for all compounds for flow rates of 1.0 or 2.0 mL min^{-1}. Details can be found in the supporting information (supplementary Table 7).

Normalization of Retention Indices

Our previous study showed that RI obtained at different initial temperatures can be transformed to those at a standard temperature by multiple linear regression which included use of molecular structural descriptors [24]. In this work, we attempted to achieve normalization of retention indices for different heating rates by use of partial least-squares (PLS) regression. The purpose of this study was to transform retention indices obtained by use of different heating rates (6, 8, and 10 °C min^{-1}) to those at a standard heating rate (4 °C min^{-1}), considering that changes of retention indices under different conditions are essentially caused by their different structures. Therefore, taking structure descriptors into consideration for normalization is reasonable. So, 59 commonly used descriptors were calculated by use of Dragon 6.0 software. The retention index at 4 °C min^{-1} was the dependent variable *y* and the retention index 6 °C min^{-1} (or 8 or 10 °C min^{-1}) plus 59 descriptors were the independent variables *X*, PLS regression was then performed. The results of normalization with descriptors, normalization without descriptors, and no normalization can be found in the supporting information (supplementary Tables 8 and 9). The error distribution is shown in Fig. 1. From the results it is apparent that normalization with descriptors is best.

Use of the RI Database for Essence Oil Analysis

Another purpose of this study was to use the created RI database for qualitative identification of aroma compounds,

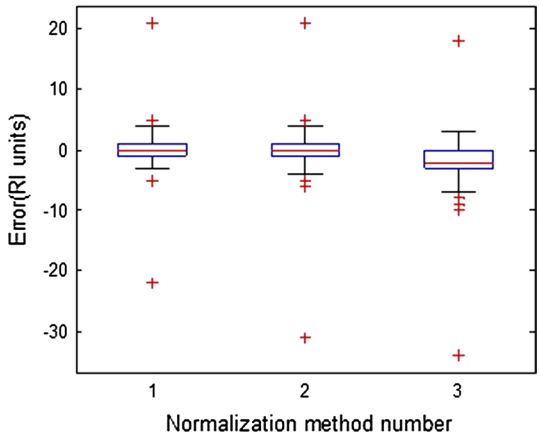

Fig. 1 Comparison of errors for different normalization methods *1* PLS normalization with descriptors, *2* PLS normalization without descriptors, *3* no normalization

so further experiments were performed to analyze the volatile fraction from a real matrix—tobacco flavor. GC–MS is often used for tobacco flavor analysis. However, a mass spectrum cannot provide enough structural information to distinguish all compounds easily, especially when there are many isomers that further increase the difficulty of quick and accurate identification. Previous studies have shown that compounds could not be distinguished by MS but could be distinguished by use of retention indices. For instance, the mass spectra of isoborneol and borneol are almost completely identical, but there is an obvious difference between their retention indices.

Although RI have been used for identification for a long time, the procedures used are rarely reported. In general, the procedure commonly used for identification by use of RI is:

1. decide which retention index will be used, e.g. Kováts index, linear retention index, or Lee index;
2. design the experiment for RI determination as similar as possible to that used for the reference RI collection used; and
3. for an ambiguous MS matching result, compare the RI calculated for a given compound with the reference RI from the reference database; the candidate with the RI closest to the calculated RI is a reliable identification.

Table 4 Identification of aroma compounds in tobacco flavor by RI and MS

Chemical Name	MS[a] (%)	$RI^{b}_{calculated}$	$RI^{c}_{database}$	$\triangle RI^{d}$
Ethyl Vanillin	95	1401	1407	-6
Pyrazine, trimethyl-	92	982	979	3
Benzoic acid	89	1150	1155	-5
1-Butanol, 3-methyl-, acetate	93	863	859	4
Butanoic acid, ethyl ester	94	786	787	-1
1-Butanol, 3-methyl-	90	724	722	2
1,2-Cyclopentanedione, 3-methyl-	94	990	994	-4
4*H*-Pyran-4-one, 2-ethyl-3-hydroxy-	92	1165	1161	4
Benzeneacetic acid, methyl ester	91	1143	1144	-1
Ethanone, 1-(4-methylphenyl)-	95	1151	1151	0
Nonanoic acid, ethyl ester	97	1273	1278	-5
Benzoic acid, 2-hydroxy-, 2-methylpropyl ester	93	1448	1444	4
Isoborneol	95	1159	1160[e]	-1
borneol	95	1166	1168[e]	-2
6-Octen-1-ol, 3,7-dimethyl-, formate	94	1252	1256	-4
D-Limonene	90	1026	1020	6
Cyclohexanone, 5-methyl-2-(1-methylethyl)-, (2*R-cis*)-	94	1127	1130	-3
Citronellol	89	1205	1208	-3
Nonanoic acid	91	1266	1260	6
Benzeneacetic acid, phenylmethyl ester	95	1793	1795	-2
2-Buten-1-one, 1-(2,6,6-trimethyl-1-cyclohexen-1-yl)-	93	1395	1390	5

[a] Results from matching of experimental mass spectra with those stored in the reference database, i.e. NIST

[b] Retention index calculated under the same experimental conditions as used to construct the RI database used

[c] The reference retention index in the homemade RI database

[d] Difference between the calculated RI and the reference RI in the database

[e] Two retention indices cited in the literature

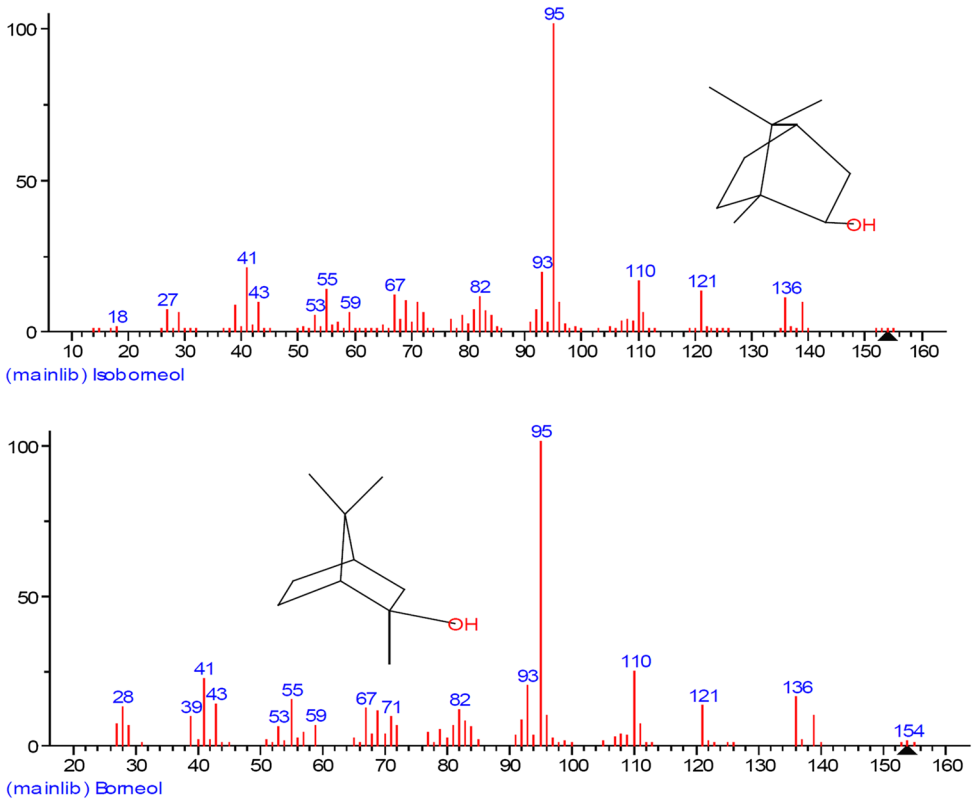

Fig. 2 MS spectra of isoborneol (*up*) and borneol (*down*)

In this work, an industrial tobacco flavor provided by the Technology Center of China Tobacco, Hunan Industrial, was used to evaluate and validate the RI database. Analysis of the tobacco flavor was performed under experimental conditions similar to those used for creation of the RI database, in accordance with the above procedure. The results are shown in Table 4. Nineteen compounds from this tobacco flavor were identified by use of RI comparison. The difference between the calculated RI and the reference RI for a given compound is quite small, the range of absolute $\triangle$RI is 0–6 RI units. The results obtained by RI comparison are in good agreement with those from matching of mass spectra.

It should be noted that the mass spectra of the two isomers isoborneol and borneol are quite similar (Fig. 2). It is very difficult to assign these peaks accurately on the basis of MS match alone. However, the two isomers elute at different times, so we can distinguish them on the basis of order of elution or retention index. On the HP-5MS column the retention indices of isoborneol and borneol were 1,160 and 1,168, respectively. So, the first peak is isoborneol and the second is borneol.

Conclusion

A database containing RI of over 300 compounds was created and used for identification of aroma compounds. Different experimental conditions (stationary phase, flow rate, heating rate, and initial temperature) were used to study their effects on RI. Stability and reproducibility of the retention index database was evaluated by replicate injection and retention regulations analysis. The data collected by us may add to the value of data collected previously. In addition, the created database was successfully used to identify aroma compounds in tobacco flavor. Further study of quantitative structure–retention relationships and data mining on the basis of this database will be conducted in the future.

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Conflict of interest We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work, there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the position presented in, or the review of, the manuscript entitled "Retention Indices for the Identification of Aroma Compounds in Temperature-Programmed Gas Chromatography: Development and Application of Retention Index Database".

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