Preparative Gas Chromatography and Its Applications

Hua-Li Zuo¹, Feng-Qing Yang¹*, Wei-Hua Huang² and Zhi-Ning Xia¹

¹School of Chemistry and Chemical Engineering, Chongqing University, Chongqing 400030, China, and ²Institute of Clinical Pharmacology, Hunan Key Laboratory of Pharmacogenetics, Central South University, Changsha 410078, China

*Author to whom correspondence should be addressed. Email: fengqingyang@cqu.edu.cn

Received 1 October 2012; revised 10 March 2013

Although hundreds of papers related to preparative gas chromatography (pGC) have been published since the late 1950s, the success of the GC technique has largely been associated with analytical instead of preparative purposes. Actually, pGC is an ideal alternative technique for the preparation of pure substances, especially volatile compounds. This paper reviews the papers (written in English) associated with pGC published over the period from the 1950s to the 2010s. For large scale preparation, large sample injection and vaporization, a high loading capacity column, a gas splitter at the end of the column and a special collecting device are fundamentally important for a pGC system. The primary components of pGC system, including injector, column, splitter, detector and collection traps, are briefly introduced. Furthermore, the applications of pGC in the separation and purification of volatile compounds from natural essential oils, in addition to the purification of isotopes, isomers and enantiomers are summarized.

Introduction

Gas chromatography (GC), invented by Martin and Synge in 1941 (1), was first applied for the separation of a series of fatty acids in 1951 (2). After this, the GC technique developed rapidly and was the first analytical instrument to be controlled by a computer. The term preparative GC (pGC) usually describes the use of large-diameter columns to purify relatively large quantities of materials by using a sample collection system at the column outlet (3). Currently, GC is one of the most important and widespread techniques in the field of analytical chemistry. However, the success of the GC technique is largely associated with chemical analysis instead of pGC (4). Although Varian Corporation commercialized a preparative scale GC (Model PSGC-10/40) late in the last century, only a preparative fraction collector for GC (Gerstel PFC, Gerstel GmbH & Co.) is still available on the market today.

Actually, pGC is an important and practical alternative to other preparative techniques such as preparative high-performance liquid chromatography (pre-HPLC), especially for volatile compounds. There have been many applications of pGC in the fields of synthetic chemistry (5–14); the fine chemical industry (15); the preparation of flavors and fragrances, or odoriferous constituents (16–19), which are primarily used in cosmetics and hygiene products; pharmaceuticals (20, 21); biology (22, 23); toxicology (24); and compound-specific radio carbon analysis (CSRA) (25, 26). In addition, the pGC system is used in the separation of large quantities of pure compounds from natural sources.

According to the SciFinder database (Figure 1), papers related to pGC have been published continuously since the late 1950s, and more than 200 papers were published during the 1960s. Although the publications related to pGC decreased during the late 20th century, pGC has regained attention by researchers during the past decade. It is believed that the characteristics and advantages of pGC will gain more attention, and it is expected that pGC will be a complementary technique for pre-HPLC in the future. Several good books or book chapters have dealt with pGC (29–31). In this paper, based on the articles (written in English) associated with pGC published over the period of 1950s to 2010s, the components of the pGC system and the applications of pGC were reviewed to provide scientific information for further studies.

pGC system

The pGC system consists of a carrier gas system (including the carrier tank, its path and a pressure controller), an injection system (including injection port and vaporizer), a separation column, splitter and detector and a trap used to collect the effluents, which was not included in the analytical equipment. Today, most of the pGC systems reported in the literature for preparative purposes have been modified from the analytical systems.

Sampling

Usually, the injection of the sample is accomplished manually by a syringe (20), which is a laborious and time-consuming process. An automated sampler controlled with a microprocessor has been used, whereas the splitless injection mode is most commonly used for preparative capillary GC (pcGC) (22, 23). Furthermore, the cool injection system (CIS), which restricts both thermal degradation and chromatographic discrimination of the analyte to some extent, has also been frequently applied in pGC and in analytical GC (33–35). In addition, a CIS contains a mass flow controlled switching device combined with a dual column system pGC, designed by Rijks and Rijks, which did well in the separation of complex samples, especially those of low concentrations (36). A newly invented injection method for pGC named a gas booster sample injection device was combined with a large volume sample dosing ring and a pneumatic six-way valve to control the sample introduction, and it also adopted high-temperature desorption combined with diaphragm booster. As a result, this unit efficiently transferred large amounts of the analyte to the sample loop (37, 38).

The loading quantity is considered to be one of the key factors that affect the separation efficiency for pGC. Therefore, special attention should be paid to sample overloading, because this can result in poor separation of the compounds or broader peaks that decrease the resolution. However, this does not mean that a...
large sample introduction is absolutely not allowed. For example, if the amount of monomer obtained from multiple small injections was less than a single larger injection, slight overloading might be acceptable. Although this also suffers from the loss of resolution, it is compensated by consuming much less time. To improve the injection amount, an expanded vaporizer is one of the most frequently used approaches. For example, for the separation of volatile components from Curry rhizome, an 8 cm × 6.60 mm vaporizer was used (39). Furthermore, Staerk et al. (40) modified the injector with an expansion vessel constructed from a 1 m × 0.53 mm i.d. stainless steel tube and connected to the system by a six-way valve. However, the large amount of injection may lead the volatile components with higher boiling points condensed in the system, which may cause a series of problems, including the blockage of conduits and joints, fluctuations in pressure and recontamination of the purified compounds. To solve this problem, Maroulis et al. (41) designed a container at the injection site to supply liquid solvent to wash away the deposit effluent after the last separation was finished (or before the following preparation started). In addition, vaporization of the sample before injection can prevent the high pressure caused by instant gasification and avoid the pollution of the injection port by the non-volatile compounds, and thus the injection amount may be improved (15).

Column

Gas-liquid chromatography (GLC) is primarily used for the preparation of low-boiling halides (42) or for the study of the structure or configuration of compounds (43–47). The preparation quantities of GLC may go from nanogram to microgram (48). Although gas-solid chromatography (GSC) allows the harvest of much larger quantities of the target compounds, it has primarily been applied for more types of different chemical compounds (39, 49) or for individual pharmacology/toxicology research. Capillary columns, belonging to GLC, have predominantly been used in GC (including pGC). However, the yielded compounds may be contaminated by the vaporized stationary phase when a high separation temperature is applied (42). Therefore, the stationary phase used in a pGC column must be physically and thermally stable, and chemically inert, to avoid taking part in chemical reaction during the preparation process (50). On the other hand, for infrared (IR) or nuclear magnetic resonance (NMR) analyses, the response of the stationary phase may mix with the compounds of interest. Therefore, the IR and NMR absorptions of 17 liquid stationary phases (including carboxylic esters, ethers and silicones) commonly used in pGC have been reported (51).

To enhance production, much effort has been devoted to increasing the loading capacity of a column by extending its diameter (52). However, increasing the column diameter can lead to extreme losses in column efficiency and separation resolution (3, 53). The efficiency of preparative columns with a relatively large diameter (up to 40 mm) is approximately half that of normal analytical packed columns (2–4 mm) (54). There is no doubt that the larger the column diameter, the slower the heat delivery rate from the outside to the center of the column (55). Therefore, the larger diameter columns can lead to inhomogeneous heating when the oven temperature is programmed to rise, which results in a retention time drift from time to time. It has also been observed that the maximum flow rate and concentration of a component occur in the center of a column (56). The efficiency of large diameter columns can be improved by increasing the length of the column or increasing the percentage of the stationary liquid, or by operating at lower or isocratic temperature. However, accompanied by a considerable increase in elution time, peak broadening may occur for compounds with a high boiling point (bp) (57). Bayer et al. (58) described a theory to explain why column efficiency was lost with greater diameter. By investigating the reproducibility of column packing, they indicated that if the columns with 5 to 10 cm diameters were packed under a longer time with consistent rocking, they may as efficient as columns with 1 cm diameter. Reiser (59) suggested an idea through an experiment that internal radial finned columns provided higher efficiency and resolution than regular columns with the same diameter. On the other hand, Weinreich (57) proposed a column with a 3-in.-thick diameter that was heated inside instead of outside, which appropriately eliminated the serious temperature gradient caused by conventional columns. A method by using analytical columns in parallel with a pGC column may either save time or avoid a decrease in

![Figure 1. Publications related to pGC according to the SciFinder database.](https://example.com/figure1.png)

![Figure 2. FID chromatogram of technical p-nonylphenol on an HP-5 MS column (30 m × 0.32 mm i.d., 0.5 μm) and selected fractionation time windows. Eleven fractions containing 77–552 μg of isomers were collected after 600 single injections (32). Adapted from Fig. 2 in Meinert, C., Moeder, M., Brack, W.; Fractionation of technical p-nonylphenol with preparative capillary gas chromatography, Chemosphere, 2007, Vol. 70, p217; Copyright © 2007 Elsevier Ltd., with permission from Elsevier.](https://example.com/figure2.png)
<table>
<thead>
<tr>
<th>Sample</th>
<th>Target compounds</th>
<th>Injection</th>
<th>Column</th>
<th>Carrier gas</th>
<th>Detector</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Methanol extract of Curcuma rhizome</td>
<td>β-Elemene, curzerene, curzerenone, curcumol and curcumone</td>
<td>Manual injection</td>
<td>Packed column with 10% OV-101</td>
<td>Nitrogen</td>
<td>FID</td>
<td>39</td>
</tr>
<tr>
<td>Peppermint oil</td>
<td>Menthol and menthione</td>
<td>Automated</td>
<td>DB-5</td>
<td>Hydrogen</td>
<td>FID</td>
<td>145</td>
</tr>
<tr>
<td>Volatile components of maize</td>
<td>Fractions attractive to Cotesia marginiventris females</td>
<td>Automated</td>
<td>On-column injection</td>
<td>Helium</td>
<td>FID</td>
<td>108</td>
</tr>
<tr>
<td>Volatile components extracted from coriander</td>
<td>(E)-2-Undecenol, 2-phenylethanol, (E,E)-2,4-dodecadien and an unknown compound</td>
<td>Automated</td>
<td>Supelco-WAX</td>
<td>Helium</td>
<td>FID</td>
<td>106</td>
</tr>
<tr>
<td>Essential oil from Acorus tatarinowii</td>
<td>cis- and trans-Aasarone</td>
<td>Manual injection</td>
<td>Packed column with 10% OV-101</td>
<td>Nitrogen</td>
<td>FID</td>
<td>49</td>
</tr>
<tr>
<td>Essential oil of Radula perrottetii</td>
<td>Viscida-3,9,14-tetraene, viscida-3,11(18), 14-tetraene, bisabol-2,6,11-tetraene,</td>
<td>Manual injection</td>
<td>Supelco-WAX</td>
<td>NA</td>
<td>NA</td>
<td>146</td>
</tr>
<tr>
<td>Essential oil of Plagiochla aspleides</td>
<td>Aromadendra-1(10),3-diene, en-4-epi-maaliol, bisabol-1,3,5,7(14)-tetraene,</td>
<td>Manual injection</td>
<td>Packed column with SE-30 on Chromosorb W-HP</td>
<td>Helium</td>
<td>FID</td>
<td>147</td>
</tr>
<tr>
<td>Essential oil of Otsostegia integrofalia</td>
<td>(E)-1-Methyl-4-(5,9-dimethyl-1-methylene-deca-4,8-diencyl)-cyclohexene</td>
<td>Manual injection</td>
<td>Packed column with SE-30 on Chromosorb W-HP</td>
<td>NA</td>
<td>FID</td>
<td>100</td>
</tr>
<tr>
<td>Essential oil of Meum athamanticum</td>
<td>α-/β-Barbatene, isobazzanene and isobarbaten</td>
<td>Manual injection</td>
<td>Packed column with OV-1701 on Chromosorb W-HP</td>
<td>Helium</td>
<td>FID</td>
<td>120</td>
</tr>
<tr>
<td>polita, Bazzania japonica and Marsupella emarginata</td>
<td>lancedoicde</td>
<td></td>
<td>1D: DB-6 column; 2D: DB-Wax</td>
<td>Hydrogen</td>
<td>FID</td>
<td>60</td>
</tr>
<tr>
<td>Mixture of peppermint, spearmint and lavender</td>
<td>Geraniol, carvone, linalyl acetate and pulegone</td>
<td>Automated</td>
<td>Packed column with SE30 on Chromosorb G-HP</td>
<td>Helium</td>
<td>FID</td>
<td>103</td>
</tr>
<tr>
<td>essential oils</td>
<td></td>
<td></td>
<td>2D-5</td>
<td>Helium</td>
<td>FID</td>
<td>114</td>
</tr>
<tr>
<td>Homalia, Plagiothecium and Taxiphyllum</td>
<td>1,4-Dimethoxybenzene</td>
<td>Automated</td>
<td>1D: DB-5 column; 2D: DB-Wax</td>
<td>Hydrogen</td>
<td>FID</td>
<td></td>
</tr>
</tbody>
</table>

*Note: NA indicates not available.
signals (70, 71). The influence of carrier gas on the separation efficiency (20), but it might be difficult to control the components simultaneously eluting from each column. Currently, the novel preparative multidimensional gas chromatography (prep-MDGC) combined with heart-cut device has demonstrated a more efficient separation, particularly for complex samples (5, 17, 22, 60–63), and a three-channel electronic pressure control module (EPC) has been used to provide sufficient pressure for the Deans switch (DS) to achieve a high separation resolution even when relatively large amounts of sample are injected (65). pcGCs equipped with cutting and back-flushing devices described by Schomburg et al. (54, 64) have been used for discarding the non-volatile material and separating the selected part in high resolution, and the pGC system composed with a set of recirculated columns designed by Golay et al. (65) can be considered to be the foundations on which prep-MDGC is laid. In addition, the use of prep-MDGC equipped with different columns makes it possible to obtain the target compounds in complex matrices by different stationary phases (66).

**Splitter and detector**

When the splitter is installed in the front of the column, split injection can be adopted. To ensure a certain volume of sample, splitless injection is commonly employed in pGC (32, 33, 61). The splitter installed behind the column allows the simultaneous detection and collection of the effluent. For destructive detectors such as the most widely used flame ionization detector (FID), the splitter separates the gas flow and diverts a small part to the detector and the rest of the flow to the traps. Furthermore, because the splitter ratios may have great effects on the detection and collection of the effluent and on the total efficiency of the pGC system (4), the splitter ratios need to be investigated in some cases (67). Roeraade and Enzell designed a pneumatically controlled splitter, which can achieve a split ratio between 0 and 100%, depending on two independently controlled regulating systems for instantaneous change (68).

With the use of a thermal conductivity detector (TCD), a non-destructive detector, it is not necessary to set the splitter after the column. However, the sensitivity of the detection can be influenced by the response time of the TCD. Modell (69) invented a backed-bed TCD with a short response time, which is less influenced by the flow rate of the carrier gas than conventional detectors. Maroulis et al. (41) designed a conduit with copper jackets that was coupled to sources of thermal energy and joined within the TCD to prevent undesired condensation within the conduits.

In the last two decades, hyphenation of GC with mass spectrometry (MS) and NMR, which are efficient methods used for structural determination, has gained increasing interest. Although GC–MS has widely been used, the MS detector is rarely applied in pGC. Nevertheless, Mandalakis and Gustafsson (34) combined a pGC system and accelerated MS (AMS) to measure the 14C contents of polycyclic aromatic hydrocarbons (PAHs).

**Carrier gas**

The choice of carrier gas is important because it affects the separation efficiency in addition to the sizes and shapes of detector signals (70, 71). The influence of carrier gas on the separation process has been discussed in detail by Verzele (72). Although various carrier gases have been used in analytical GC (71), pGC only adopts the three most commonly used gases, including hydrogen, nitrogen and helium. Nitrogen is a more suitable carrier gas for pGC than hydrogen and helium, because of the high price of helium and the potential danger of hydrogen. In addition, the flow rate of carrier gas also affects the sensitivity of TCD (69) and the trapping efficiency (73), and an overly high flow rate may flush away the liquid stationary phase (50, 51).

**Trap (fraction collector)**

The collection of the fractions eluted from pGC is as important as the separating process (74). The efficiency of a trap used to collect the target effluent may simply be expected to be as high as possible. However, affected by various factors, recovery values of the selected eluates may be low and fluctuating. Poor trapping efficiency is sometimes caused by the formation of aerosol (75–77), especially for those components with poor volatility. It has been demonstrated that the recovery values were proportional to the volatility of target compounds, and the temperature of the collecting device also intensely affected the efficiency (73). It is imperative that the temperature of the connecting channel between the collecting device and the GC system be maintained above the bp of the constituents to prevent undesired condensation (20). Many conventional methods for the collection of samples from pGC are available, including thermal gradient traps (78–80), electrostatic precipitation (81–83). Volman collector (84) with cartridge or aluminum rod heater and fritted filter traps (77), which all provide efficient recovery values of aerosol forming compounds. Furthermore, the trapping method of effluent in a glass coil immersed in liquid nitrogen with a vacuum manifold device can also effectively avoid the aerosol formation and sample losses (85). In addition, a trap using potassium bromide (KBr) powder as an inert support to adsorb the sample can simplify the following IR identification step (86).

Actually, cold traps, on which many modified traps are based, are most frequently used in pGC (77, 87). The temperatures for the cool traps are closely related to the bp of the target compounds. For compounds with high bp, room temperature
circumstances can serve as adequate cooling conditions; for compounds with moderately low bp, solid CO$_2$ in a Dewar flask is preferred (88); for compounds with extremely low bp, liquid nitrogen should be employed (34, 89, 90). A pear-shaped flask (91) or a trap consisting of a gas dispersion tube (bubbler) with a fritted cylinder (92) may improve the recovery to some degree. Furthermore, for collecting compounds with high molecular weight such as methyl esters of the polygenic acids and fatty acid methyl esters, which more easily lose collecting efficiency, a particular trap was established, consisting of a series of bottles fitted with sintered glass distributors and containing acetone connected in series with the outlet (93). In addition, Roeraade and Enzell designed a six-port flow distributor made of platinum-iridium at the end of the column. The multi-channel distributor connected to the traps consisted of coiled glass capillary tubing, the inner walls of which were wetted with suitable solvent and cooled in liquid nitrogen; this ensured maximum trapping efficiency, even for very small amounts of material (68). In recent years, some new styles of collectors have been introduced. Liu et al. designed an efficient trap that consists of an enrichment unit with a temperature controller, a second diaphragm pump (differing from that in the sampling system), a gas

Figure 4. FID chromatogram for asarone isomers on a stainless steel column packed with 10% OV-101 (3 m x 6 mm i.d.) and selected fractionation time windows (A); capillary GC–FID chromatogram of the collected fractions (B); MS spectra of the collected fractions (C); 178 mg and 82 mg, respectively, of cis-asarone (F1) and trans-asarone (F2) were collected after 90 single injections (5 μL) (49). Adapted from Figures 1 and 2 in Zuo, H.L., Yang, F.Q., Zhang, X.M., Xia, Z.N. Separation of cis- and trans-asarone from Acorus tatarinowii by preparative gas chromatography; Journal of Analytical Methods in Chemistry, 2012, vol. 2012, p2; Copyright © 2012 H. L. Zuo et al., with permission from the authors.
component collection tube and a cold well (94). The inlet of the enrichment unit is connected to the outlet of the chromatographic detector via a valve. Hu et al. tried to make a preparative fraction collector with improved reliability and automation by using a microprocessor controller and a corrosion resistance magnetic valve (95). Moreover, traps based on megabore capillary columns, named open tubular traps (OTTs), have also been used in pGC (96, 97).

Currently, the commercially available fraction collector for GC (Gerstel PFC), equipped with six sample traps and one waste trap, collects individual fractions after GC separation. The PFC system is controlled by a microprocessor, which automates the collection process. Although the PFC has been used in several studies (23, 98, 99), the technical limitation to only six collectable fractions using one PFC still hinders its applications. One study combined two PFCs via a special zero-dead volume effluent splitter, which enhanced the number from six to 12 collectable fractions within one GC running cycle (32). This method provided the fractionation of nonylphenol isomers into 11 fractions containing 77–552 µg of isomers collected after 600 single injections (Figure 2). This yield is sufficient to allow subsequent bio-testing in the E-screen assay. Therefore, a PFC system developed with more sample traps can effectively enhance its applications, especially for those samples with multiple components.

Applications of pGC

pGC is widely used in various fields, either to obtain an individual compound for structural identification or to produce a certain amount of pure compound for application in industrial large-scale preparation. In laboratories, pGC has been a practical tool to prepare small quantities of high purity materials.

Volatile components from natural essential oil

Essential oils are some of the most valuable natural products with multiple pharmacological activities. pGC is frequently used to collect desired fractions of the effluent from many other naturally present matrix components, and has been regarded as an ideal approach to separate volatile components from natural materials. The applications of pGC for separating volatile components from natural essential oil are summarized in Table 1.

The volatile compounds of essential oils from natural sources have primarily been extracted by hydro-distillation (100–104), reflux heating with less polar organic reagent (104) or a water–alcohol system (105). Because a moist sample can damage the column, it is better to remove the water that is infused during the extraction process, e.g., by drying over anhydrous sodium sulfate (106, 107). On the other hand, the sample subjected to a GC system must first be thermally stable enough to avoid thermal degradation (20), and of course, the sample should be vaporizable because the non-volatile compounds can pollute the GC system. Furthermore, the pGC system adopts larger diameter columns and larger loading sample size, which will undoubtedly result in a large decrease in the plate number and separation resolution. Thus, proper pretreatment of the sample is needed to produce the desired purity of products. The pGC preparation procedure can be simplified by combining with other techniques such as separation and removal of the unconcerned components by silica gel chromatography (8, 49) or collection of the stripping components adsorbed by the silica medium (108, 109), extraction of the target components by using a certain polar reagent or collection of the distilled product under a certain temperature range (110). On the other hand, several non-volatile or semi-volatile compounds can contaminate the injection port and the column head if the crude oil is directly injected into GC system. Therefore, removing the non-volatile components by silica gel chromatography can extend the lifetime of the system (39).

Low volatility compounds such as terpenes, sesquiterpenes and sesquiterpenoids, which are ubiquitous in essential oil, are usually isolated by pGC (39, 101, 102, 107, 111, 112). Staritas et al. (103) obtained four terpenes, 15 sesquiterpenoids, two diterpenoids and the aliphatic metabolites from the hydrodistillation product of moses by pGC. β-Elemene, curzerene, curzerenone, curcumol and curcumene (with yields of 5.1–46.2 mg after 83 single injections of 20 µL) were separated

<table>
<thead>
<tr>
<th>Sample</th>
<th>Target compounds</th>
<th>Injection</th>
<th>Column</th>
<th>Carrier gas</th>
<th>Detector</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mushroom odor</td>
<td>1-Octenyl-3-acetate</td>
<td>Manual injection</td>
<td>Packed column with modified β-CD on Chromosorb P-NAW</td>
<td>Hydrogen</td>
<td>TCD</td>
<td>128</td>
</tr>
<tr>
<td>Mixture of enflurane and isoflurane</td>
<td>Enflurane and isoflurane</td>
<td>Manual injection</td>
<td>Packed column with SE-54 on Chromosorb P-NAW DMCS</td>
<td>Helium</td>
<td>FID</td>
<td>129</td>
</tr>
<tr>
<td>Racemic 2-chloropropionate</td>
<td>Enantiomers of methyl 2-chloropropionate</td>
<td>Six-way valve injection</td>
<td>Packed column with β-CD on Chromosorb A</td>
<td>Hydrogen</td>
<td>FID</td>
<td>40</td>
</tr>
<tr>
<td>Natural gas</td>
<td>Isotope of nitrogen</td>
<td>Direct infusion</td>
<td>NA</td>
<td>Helium</td>
<td>TCD</td>
<td>148</td>
</tr>
<tr>
<td>Racemic isoflurane</td>
<td>Enantiomers of isoflurane</td>
<td>Manual injection</td>
<td>Packed column with β-CD on Chromosorb W-AW γ-CD coated capillary column</td>
<td>Hydrogen</td>
<td>TCD</td>
<td>125</td>
</tr>
<tr>
<td>Fluoroether anesthetics</td>
<td>Desflurane, enflurane and isoflurane</td>
<td>Manual injection</td>
<td>Packed column with Pd deposited on porous α-Al2O3</td>
<td>Helium</td>
<td>TCD</td>
<td>124</td>
</tr>
<tr>
<td>Hydrogen isotope mixture</td>
<td>Triterpene and deuterium</td>
<td>Pressurized injection</td>
<td>Packed column with TBOMS-β-CD and SE-54 on Chromosorb P-NAW DMCS</td>
<td>Nitrogen</td>
<td>TCD</td>
<td>150</td>
</tr>
<tr>
<td>Racemic perhydrotriphenylene</td>
<td>All trans-perhydrotriphenylene</td>
<td>Manual injection</td>
<td>Packed column with OV-101 impregnated on Chromosorb W-AW-DMCS</td>
<td>Nitrogen</td>
<td>NA</td>
<td>151</td>
</tr>
<tr>
<td>Racemic 1-chloro-2,2-dimethylaziridine</td>
<td>(++)-1-Chloro-2,2-dimethylaziridine</td>
<td>Manual injection</td>
<td>Packed column with TBDMS-β-CD on Chromosorb W-AW-DMCS</td>
<td>BPX5</td>
<td>Helium</td>
<td>33</td>
</tr>
<tr>
<td>1,1,1-Trichloro-2,2-bis(p-chlorophenyl) ethane</td>
<td>Chlorine isotope</td>
<td>Coated injection system</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Note: NA indicates not available.*
<table>
<thead>
<tr>
<th>Sample</th>
<th>Target compounds</th>
<th>Injection</th>
<th>Column</th>
<th>Carrier gas</th>
<th>Detector</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sweet potatoes</td>
<td>Ipomeamarone, 2-Para-aryl-substituted 1,3,5-triphenylbenzene</td>
<td>Manual injection</td>
<td>Packed column with SE-30 on GC PGC</td>
<td>Hydrogen</td>
<td>TCD</td>
<td>152</td>
</tr>
<tr>
<td>Products arising from reaction of phenylacetylene with para-substituted aryllodides</td>
<td>α-Nerol and α-geraniol</td>
<td>Automated splitless injection</td>
<td>DB-5</td>
<td>Hydrogen</td>
<td>FID</td>
<td>5</td>
</tr>
<tr>
<td>Chemical reaction product</td>
<td>Tetramethyilsilane</td>
<td>Manual injection</td>
<td>Packed column with SE 30 on Chromosorb A</td>
<td>Helium</td>
<td>NA</td>
<td>153</td>
</tr>
<tr>
<td>Crude Si(CH3)4 containing n-butane, 2-methylpentane, 3-methylpentane and tetrahydrofuran</td>
<td>2,5-Dimethyl-4-hydroxy-3(2H)-furane</td>
<td>On-column injection</td>
<td>Packed column with G.E. SP96 on firebrick (1); Carbowax 20M on Chromosorb W (2)</td>
<td>Helium</td>
<td>NA</td>
<td>109</td>
</tr>
<tr>
<td>Distillate of pineapple juice</td>
<td>Juvenile hormone III, Active fraction of polybrominated biphenyl mixture</td>
<td>Manual injection</td>
<td>Packed column with OV-1 (1) or OV-17 (2) on Chromosorb W-AW-DMCS</td>
<td>Helium</td>
<td>FID</td>
<td>155, 156</td>
</tr>
<tr>
<td>Mixture of juvenile hormones</td>
<td>α-Elemene and β-bourbonene, Metal TPM complexes</td>
<td>On-column injection</td>
<td>Packed column with HP-1</td>
<td>Helium</td>
<td>TCD</td>
<td>159</td>
</tr>
<tr>
<td>Extracts of gum haggar and gum myrrh</td>
<td>Triglyceride, Off-flavor compounds in water</td>
<td>Manual injection</td>
<td>Packed column with JXR (1) or SE-30 (2) on Chromosorb W</td>
<td>Helium</td>
<td>FID</td>
<td>135</td>
</tr>
<tr>
<td>Atmospheric aerosol</td>
<td>Naphthalene, phenanthrene, fluoranthene, pyrene, chrysene and benzo(a)pyrene</td>
<td>Coated injection system</td>
<td>Capillary column coated with a cross-bonded methyl silicone</td>
<td>Helium</td>
<td>FID</td>
<td>140</td>
</tr>
<tr>
<td>Mixture of PAHs</td>
<td>Fluorene, retene, chrysene, pyrene, fluoranthene and perylene</td>
<td>Coated injection</td>
<td>Supelco SPB-5 capillary column</td>
<td>Helium</td>
<td>MSD</td>
<td>158</td>
</tr>
<tr>
<td>Portuguese wines from the Douro region</td>
<td>2,6,6-Trimethylcyclohex-2-ene-1,4-dione and diacetyl</td>
<td>Manual injection</td>
<td>Packed column with Carbowax 20 M on Chromosorb W</td>
<td>Helium</td>
<td>FID</td>
<td>159</td>
</tr>
<tr>
<td>Cod liver oils</td>
<td>3-Hopropenyl-2,2-dimethylcyclobutylmethyl</td>
<td>Manual injection</td>
<td>Packed column with SE-30 Chromosorb W</td>
<td>Nitrogen</td>
<td>FID</td>
<td>161</td>
</tr>
<tr>
<td>Petroleum distillates</td>
<td>Nortriptyline, p-hydroxybiphenyl</td>
<td>Manual injection</td>
<td>Packed column with UCW 98 on Diatoport 5</td>
<td>Helium</td>
<td>TCD</td>
<td>162</td>
</tr>
<tr>
<td>Cognac</td>
<td>Volatile components in wine</td>
<td>Manual injection</td>
<td>Packed column with Aperion L on Universal B</td>
<td>Helium</td>
<td>FID</td>
<td>163</td>
</tr>
<tr>
<td>Calvados and cognac</td>
<td>Amino acids</td>
<td>Manual injection</td>
<td>Packed column with OV-1 on Supelcoport</td>
<td>Helium</td>
<td>TCD</td>
<td>164</td>
</tr>
</tbody>
</table>

*Note: NA indicates not available; MSD indicates mass selective detector.*
from the methanol extracts of *Curcuma rhizome* pretreated by silica gel chromatography (Figure 3) (39). The *cis* and *trans*-asarone isomers (with yields of 178 and 82 mg, respectively, after 90 single injections of 5 μL) were separated by pGC from their mixture, which was fractionated from the essential oil of *Acorus tatarinowii* by silica gel chromatography (Figure 4) (49). β-Sinensal was separated from California cold-pressed orange oil by a combination of vacuum distillation, silica gel chromatography and pGC (113). Rodin *et al.* (109) used pGC to separate volatile flavor and aroma components from pineapple. Furanocoumarins alcohol was isolated from the essential oil of the liverwort *Lophocolea heterophylla* (114).

**Separation of isotopes, isomers and enantiomers**

Dating back to the 1960s, pGC has intensively been used in academia to purify the compounds arising from chemical reactions for further structural elucidation (5, 115–117). More importantly, pGC has successfully been applied for the separation of stereoisomer reaction products of synthesis or hydrolysis (43–46, 118), which represented great contributions to the research of mechanisms for chemical reactions. Furthermore, pGC also allowed the separation of diastereoisomers (119) and widespread isomers in natural essential oils (49, 120). On the other hand, although cryogenic distillation has been known and used for the separation of the hydrogen isotopes tritium and deuterium, pGC is more suitable and particularly applicable for the isolation of hydrogen isotopes on a large scale (121, 122). Furthermore, the pGC system at Joint European Torus (JET), which is a type of displacement chromatography that uses palladium (Pd) as working material because of its large isotope effects on hydrogen absorption and hydride formation (123), can produce deuterium and tritium of high purity at low temperature with good production efficiency (124).

Various cyclodextrin-based stationary phases have been used in GC for approximately 20 years (125, 126). Derivatized cyclodextrins (CDs) as chiral selectors can be used as reliable stationary phases for the pGC separation of enantiomers of different structural characteristics (127–130); this separation mechanism has been thoroughly illustrated (131). Considering the large experimental effort needed for the optimization of those preparative-scale separations (126), Staerk *et al.* (132) developed a model of elution band profiles in the pGC separation of enantiomers based on the equilibrium-dispersive model of non-linear GC, which may be used to speed the optimization process by computer modeling.

The applications of pGC for isotopes, isomers and enantiomers are summarized in Table II.

**Miscellaneous**

The separation of a few milligrams of pure substance from a mixture of biological origins is often important for the studies of steroids, alkaloids and many other compounds. A pGC method was developed for the separation of steroids and alkaloids at milligram-scale by using relatively thin-film column packing (133). Furthermore, certain quantities of low concentration fatty acids have successfully been prepared by pGC (134). The fractionation of high molecular weight triglycerides by GLC and thin-layer chromatography (TLC) on silica gel impregnated with silver nitrate has resulted in considerable progress and is widely used in the structural elucidation of natural fats and oils (135–139). On the other hand, cannabinoids, tetrahydrocannabinol and cannabidiol, which are three primary cannabinoids, have been isolated by pGC from the enriched extract of cannabis (104). As a purification technique, pGC has been used for the purification of a styrene monomer from its commercial products (140). In addition, it has been shown that wire mesh packing as carrier of the stationary phase is effective for separating the binary azetrope of 1-propanol–water (15). pGC was also used to purify metals as their volatile derivatives (141) and as a purification procedure in toxicology. It can obtain large enough amounts of toxic agents with sufficient purity from tissue for further identification (142). Miscellaneous applications of pGC are summarized in Table III.

**Conclusions**

Although pGC has widely been used, it is still not comparable to pre-HPLC, which has constantly been used in the pharmaceutical industry for the isolation and purification of biological active substances. For pGC, several aspects should be intensively improved. First, the scaled-up pGC column requires large volumes of carrier gas to provide a certain pressure, and it is difficult to reutilize the mobile phase. Second, the sample must be fully vaporized onto the column to ensure radial distribution of the sample across the column. Third, because the target compounds are eluted in a very dilute form from the column, they must be extracted or condensed from the gas stream, which makes it difficult to achieve high efficiency. Finally, although a large GC column provides a high productivity, it is difficult to achieve the goal of automatic production control; the capillary column is unsuitable for industrial applications. Nevertheless, pGC is successful in many rather special applications. For example, pCGC, combined with a computer-based structural generation tool used for mutagenicity prediction, has been applied within effect-directed analysis (EDA) to reduce sample complexity and to identify candidate mutagens in the samples (23). Since Eglinton *et al.* (143) first reported the isolation of monomer compounds by pGC and subsequent radiocarbon (14C) isotope analysis by offline AMS, the combined use of these techniques has provided a powerful analytical tool to achieve CSRA (144). Although the preparation process of CSRA can be accomplished by either HPLC or pGC, the clear and highly resolved analytical separations of the capillary columns has made pCGC more attractive for natural abundance CSRAs (22).

**Acknowledgments**

This work was supported by the National Natural Science Foundation of China (21275169, 81202886 and 21175159), the International Cooperation Project of Ministry of Science and Technology (2010DFA32680) and the Natural Science Foundation Project of CQ CSTC (2010BB5070). We are grateful to Dr. Liya Ge from Nanyang Technological University for her help in checking the written English of the manuscript.
References


2. James, A.T., Martin, A.J.P.; Gas-liquid partition chromatography; the separation and micro-estimation of volatile fatty acids from formic acid to dodecanoic acid; Biochemical Journal, (1952); 50: 679–690.


10. Los, M., Mighell, A.D.; The acid-catalysed rearrangement of 4,4a,5,6,7,8-hexahydro-5-hydroxy-1(3H)-naphthalenone; Tetrabedron (1965); 21: 2297–2303.


12. Lee, C.C., Kruger, J.E., Wong, E.W.C.; On the problem of 1,3-hydrate shifts during nitrous acid deamination of 1-propylammonium perchlorate; Journal of the American Chemical Society; (1965); 87: 3985–3986.

13. Tsuji, T., Morikawa, M., Kiji, J.; Organic syntheses by means of noble metal compounds X. Carboxylation reaction of cyclopropane catalyzed by palladium chloride; Tetrabedron Letters; (1965); 6: 817–819.


17. Marriott, P.J., Eyres, G.T., Dufour, J.P.; Emerging opportunities for flavor analysis through hyphenated gas chromatography; Journal of Agricultural and Food Chemistry; (2009); 57: 9962–9971.


23. Meinert, C., Schymanski, E., Küster, E., Köhne, R., Schürmann, G., Brack, W.; Application of preparative capillary gas chromatography (pGC); automated structure generation and mutagenicity prediction to improve effect-directed analysis of genotoxins in a contaminated groundwater; Environmental Science and Pollution Research; (2010); 17: 885–897.


32. Meinerz, C., Moeder, M., Brack, W.; Fractionation of technical α- and γ- n-nonylethylphenol with preparative capillary gas chromatography; Chemosphere; (2007); 70: 215–223.


34. Mandalakis, M., Gustafsson, Ö.; Optimization of a preparative capillary gas chromatography-mass spectrometry system for the isolation and harvesting of individual polycyclic aromatic hydrocarbons; Journal of Chromatography A; (2003); 996: 163–172.


42. Brazhnikov, V.V., Sakodynskii, K.I.; The purification of germanium tetraiodide by preparative gas chromatography; *Journal of Chromatography A*, (1972); 64: 157–161.

43. Nagai, M., Tanaka, O., Shibata, S.; The stereochemistry of protopanaxadiol the absolute configuration of C(20) of dammarenelone-1 and-2; *Tetrahedron Letters*, (1966); 7: 4797–4801.

44. Park, J.D., Cook, E.W.; Sterechemical aspects of nucleophilic substitution at unsaturated fluorocarbon; *Tetrahedron Letters*, (1965); 4: 4853–4855.

45. Lim, D., Kolimsky, S., Štokr, J., Petriček, J.; Separation and characterization of stereoisomers of 2, 4, 6-trichlorohexane; *Journal of Polymer Science Part B*, (1966); 4: 577–579.


106. Ikeura, H.; Identification of (E, E)-2, 4-Undecadien-1-al from Coriander (*Coriandrum sativum* L.) as a highly effective deodorant compound against the offensive odor of porcine large intestine; *Journal of Agriculture and Food Chemistry* (2010); 58: 11014–11017.


117. Meijnald, J., Gassman, P.G., Hurst, J.J.; Highly strained bicyclic systems. VIII. The nitric acid deamination of 5, 5-dimethylbicyclo-[2.1.1]-hexyl-2-β-amine; *Analytical Chemistry* (1962); 84: 3722–3726.


127. Schurig, V.; Use of derivatized cyclodextrins as chiral selectors for the separation of enantiomers by gas chromatography; Annales Pharmaceutiques Francaises, (2010); 68: 82–98.


137. Breckenridge, W.C., Kuksis, A.; Structure of bovine milk fat triglycerides; Lipids (1975); 13: 497–505.


147. Adio, A.M., König, W.A.; Sesquiterpene constituents from the essential oil of the liverwort Plagiochila asplenoides; Phytochemistry; (2005); 66: 599–609.


156. Mahmood, F., Carlson, D.A., Borovsky, D.; Biosynthesis and metabolism of juvenile hormone III from methyl farnesoate by exposed corpora allata of Lutzomyia antennipalpis; Journal of Medical Entomology; (1992); 29: 548–555.


158. Boger, F.S.S., Castro, H., Fortunato, N., Azevedo, Z., Macedo, A., De Freitas, V.A.P.; Chemicals with sweet aroma descriptors found in Portuguese wines from the Douro region: 2, 6, 6-Trimethylcyclohex-2-ene-1,4-dione and diacetyl; Journal of Agricultural and Food Chemistry; (2001); 49: 263–269.


162. Lochte, H.L., Pittma, A.G.; Nitrogen compounds of petroleum distillates. XXVIII. Isolation of 2-methyl-6,7-dihydro-5H-1-pyridine; Preparation of some methylhydropryridines; Journal of the American Chemical Society, (1960); 82: 469–472.
