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Synthesis and pyrolysis of two flavor precursors of oct-1-en-3-yl methylpyrazinecarboxylates

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Abstract To rich flavor additive species of pyrazines, two new compounds of 3,6-dimethyl-2,5-pyrazinedicarboxylic acid 1-octen-3-yl ester (DMPOE) and 3,5,6-trimethyl-2-pyrazinecarboxylic acid 1-octen-3-yl ester (TMPOE) were synthesized by $KMnO_4$ oxidation, acylating chlorination and esterification reaction, in which tetramethylpyrazine and 1-octen-3-ol were used as initial materials. Thermogravimetry (TG), differential scanning calorimeter (DSC) and pyrolysis–gas chromatography/mass spectrometry (Py–GC/MS) were conducted to investigate the thermal degradation behaviors of DMPOE and TMPOE. TG–DTG results indicated that the T_p of DMPOE and TMPOE with the largest mass loss rate was at 310 and 250 °C, respectively. The T_{peak} of DMPOE and TMPOE showed by DSC curves was 301.8 and 260.0 °C, respectively. Py–GC/MS was performed to benefit the simulation of cigarette burning conditions, and the results indicated that DMPOE and TMPOE could release specific flavors of 1-octen-3-ol and diversified alkylpyrazines. Furthermore, the thermal degradation mechanisms of the flavor precursors of DMPOE and TMPOE were discussed. The study on the thermal behavior of these two methylpyrazinecarboxylates would provide theoretical basis for their application in tobacco.

Keywords Pyrazines · 1-Octen-3-ol · TG · DSC · Py–GC/MS · Pyrolysis

Introduction

Pyrazines are heterocyclic aromatic organic compounds with the underlying chemical formula $C_4H_4N_2$, which are commonly used in medicine, organic chemistry, food and materials [1]. Pyrazines are considered to provide a representative roasted flavor in foods and commonly exhibit the sensory properties of toasted and nutty odor notes. Because of their unique organoleptic properties, they have recently been receiving increased attention in food-related areas, and their concentrations detected in foods are in the range from approximately 0.001 to 40 ppm. They have been found in cocoa beans, grilled beef, potato, kohlrabi, wheat bread, asparagus and liquors, which are toasted, roasted or fermented in their preparation, or that involve an extended heat treatment during the isolation procedure [2–6]. These pyrazines are usually formed from a series of complex chemical reactions, known as the Maillard reaction or nonenzymatic browning reaction [7–11]. In the early days, numerous pyrazines have been synthesized and promoted as flavoring agents [12]. Pyrazine esters were reported as aroma precursors in tobacco field. For instance, methyl 2-pyrazinyl-2-propanyl oxalate could release 2-isopropenylpyrazine flavorant. 3,7-Dimethyl-2,6-octadien-1-yl 3,5,6-trimethyl-2-pyrazineacetate produced tetramethylpyrazine and the correspondent olefin by cracking reaction during pyrolysis. As is well-known that 1-octen-3-ol being used for the preparation of the esters spices is characterized by lavender smell and mushroom fragrance [13]. Because of its inherent flavor, 1-octen-3-ol serves as an additive to food stuffs, medicines and tobacco.

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[14, 15]. However, the volatility and low odor threshold limits its application in the high-temperature processing [16]. Over recent years, increasing attention has been paid to pyrazine ester precursors to achieve the purpose of flavor stable release, especially for high-temperature processing production. Therefore, in order to get the two characteristic flavor of pyrazines and 1-octen-3-ol, using tetramethylpyrazine and 1-octen-3-ol as raw materials, new flavor precursors of 3,6-dimethyl-2,5-pyrazinedicarboxylic acid 1-octen-3-yl ester (DMPOE) and 3,5,6-trimethyl-2-pyrazinecarboxylic acid 1-octen-3-yl ester (TMPOE) were synthesized by chemical methods. Although the two pyrazine esters are nonvolatile and flavorless, pyrolysis of them could release compounds that may contribute to flavor either directly or by subsequent rearrangement.

Thermogravimetry (TG), differential scanning calorimeter (DSC) and pyrolysis–gas chromatography/mass spectrometry (Py–GC/MS) are useful tools for studying the pyrolysis behavior of isolated tobacco additives and flavor precursors [17–20]. The researches on the thermal behavior and decomposition mechanism of compounds were reported a lot, such as materials [21], polymers [22–24] and proteins [25]. The two new compounds of DMPOE and TMPOE were rarely reported in the literature before, let alone the thermal behavior of them. Generally, the study on pyrolytic behavior of the sample is a first step in its total toxicological assessment. The aim of this research is to present the preliminary results obtained by applying TG, DSC and Py–GC/MS for the pyrolysis behavior of alkylpyrazine esters (DMPOE and TMPOE). This will provide more important basic theory for the possibility of DMPOE and TMPOE being used as tobacco flavor additives in burning processing.

Firstly, DMPOE and TMPOE were synthesized and they were characterized by proton nuclear magnetic resonance spectroscopy (^{1}H NMR, ^{13}C NMR), infrared spectroscopy (IR) and high-resolution mass spectroscopy (HRMS). Then the thermal changes of DMPOE and TMPOE in pyrolysis process were investigated by TG and DSC. According to the studies of Stotesbury et al. [26] and Baker et al. [27, 28] on the pyrolytic behavior of tobacco ingredients, we know that the temperature and pyrolysis atmosphere are the key controllable variables in experiments to mimic smoking. Inside the cigarette burning zone, there are about 9% oxygen concentration distributing at 300–700 °C and almost no oxygen at above 700 °C. Therefore, pyrolysis conditions were chosen to simulate the cigarette burning process, which were set at 300, 450 and 600 °C in atmosphere of nitrogen and 9% oxygen in nitrogen, and at 750 and 900 °C in nitrogen only. Py–GC/MS was extensively carried out for the qualitative and quantitative analysis of pyrolysis products. The thermal stability, pyrolysis products and degradation mechanism of DMPOE and TMPOE were discussed in this paper.

Experimental

Materials

DMPOE and TMPOE were prepared by oxidation, acylating chlorination, and esterification reaction (Scheme 1) using tetramethylpyrazine (99%, Sigma-Aldrich) and 1-octen-3-ol ($\geq$98%, AR) as initial materials. 1-Octen-3-ol was purchased from Henan Xinzheng Gold Leaf Spice Co., Ltd. (China). Other reagents were of analytical grade quality and purchased from Tianjin Kermel Chemical Reagent Co., Ltd (China) and, when necessary, were purified and dried by standard methods.

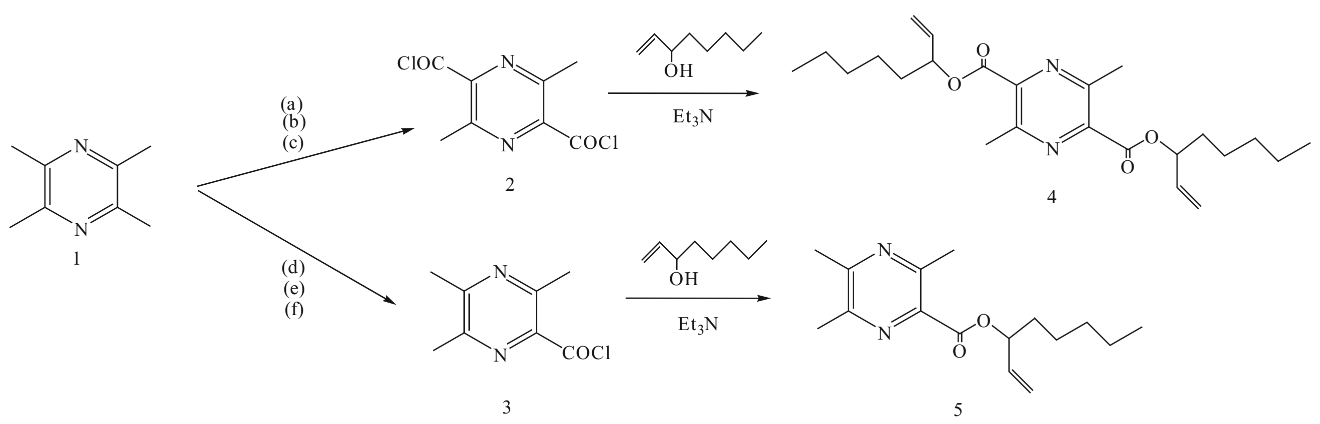
Thin-layer chromatography (TLC) was performed on silica gel GF254 (Merck), and spots were visualized by irradiation with UV light (254 nm). Silica gel column chromatography was carried out using a glass column (3 cm i.d. × 60 cm) filled with silica gel (0.03-0.06 mm, Qingdao ocean chemistry factory, China).

Preparation of DMPOE

General procedures for the synthesis of DMPOE were as follows. It was prepared according to our previous work [29], but with some modifications in the last process. 3,6-Dimethyl-2,5-pyrazinedicarbonyl chloride (compound 2) was prepared from oxidation with $KMnO_4$ at 100 °C and from acylating chlorination with $SOCl_2$, which was used directly in the next step without any purification. Compound 2 (4.2 mmol) was dissolved in 25 mL of ether. Then 0.897 g 1-octen-3-ol (7.0 mmol) and 1.0 mL (about 7.0 mmol) of dried triethylamine (Et_3N) were added to the solution of compound 2. The reaction mixture was stirred overnight at room temperature. Progress of the reaction was monitored by TLC with petroleum ether–ethyl acetate (15:1, v:v). Upon completion of the reaction, the mixture was washed in sequence with 3 × 25 mL of saturated sodium bicarbonate, 3 × 25 mL of H_2O, and 3 × 25 mL of saturated sodium chloride. The organic layer was dried (Na_2SO_4), filtered and concentrated. The residue was separated on silica gel column chromatography with petroleum ether–ethyl acetate (45:1, v:v) and recrystallized from ethyl acetate to give compound 4 (DMPOE) as white solid with purity of $\geq$98% (0.978 g, yield 67%). The yield was calculated by the last step reaction.

Preparation of TMPOE

TMPOE was prepared with some modifications according to the literature [30]. 3,5,6-Trimethylpyrazine-2-carboxylic acid was prepared from oxidation with $KMnO_4$ at 65 °C for 24 h and then underwent acylating chlorination with



Scheme 1 Synthesis of the flavor precursors of oct-1-en-3-yl methylpyrazinecarboxylates 4 (DMPOE) and 5 (TMPOE). (**a**) $KMnO_4$, H_2O, 100 °C; (**b**) H_2SO_4, pH 2–3; (**c**) $SOCl_2$, reflux; (**d**) $KMnO_4$, H_2O, 65 °C; (**e**) HCl, pH 2–3; (**f**) $C_2Cl_2O_2$, rt

$C_2Cl_2O_2$ to give 3,5,6-trimethylpyrazine-2-carbonyl chloride (compound 3). Then 0.448 g 1-octen-3-ol (3.5 mmol) and 0.5 mL (about 3.5 mmol) of dried triethylamine (Et_3N) were added to the solution of compound 3 (4.2 mmol) in 25 mL of dried CH_2Cl_2. Progress of the reaction was monitored by TLC with petroleum ether–ethyl acetate (5:1, v:v). The reaction mixture was stirred at room temperature for 3 h. On completion of the reaction, the mixture was also washed, dried (Na_2SO_4), filtered and concentrated by the same operations as above. The residue was purified twice by silica gel column chromatography with petroleum ether–ethyl acetate (20:1, v:v) to afford compound 5 (TMPOE) as colorless liquid with purity of $\geq$98% (0.537 g, yield 56%). The yield was calculated by the last step reaction.

Structure identification techniques

IR spectra were recorded with Thermo/Nicolet Nexus 470 FT-IR ESP spectrometer (USA). The samples were analyzed as KBr micropellets. ^1H NMR and ^{13}C NMR spectra data were recorded by a BRUKER AVANCE III 400 MHz spectrometer (Switzerland), and each sample was dissolved in $CDCl_3$ with tetramethylsilane (TMS) as the internal reference standard. HRMS data were obtained using the Waters Micromass Q-TOF Micro TM high resolution mass spectrometer (USA). Purity values were detected by Waters 2695 HPLC with 2996 diode array detector and chromatographic column of Agilent ZORBAX SB-C_{18} (4.6 mm × 250 mm, 5 μm).

Thermal analysis

Thermal decomposition of DMPOE and TMPOE was compared by TG-DTG and DSC analysis using a simultaneous thermal analyzer (NETZSCH STA 449 F3, Germany). The mass of each sample loaded into the alumina crucibles for each run was about 5 mg. The tests were carried out under argon atmosphere with a flow rate of 60 mL min^{-1}. Thermal degradation temperature ranged from 30 to 900 °C at a constant heating rate of 10 °C min^{-1}. The TG curve, DTG curve and DSC curve of samples were simultaneously performed.

Py–GC/MS analysis

Py–GC/MS analysis used in this study has a Pyroprobe 5250T (CDS, Analytical Inc.,) connected directly to a GC/MS (Agilent, 7890A/5975C). About 0.20 mg of each sample was centered in a 25-mm quartz tube and pyrolyzed isothermally at the designed temperatures for 10 s. The pyrolysis temperatures were set up at 300, 450, 600, 750 and 900 °C, respectively. The temperature of the reactor was initially set at 50 °C and heated nominally at the rate of 10 °C ms^{-1}. The pyrolysis atmospheres were both in nitrogen (0% oxygen) and in the mixture gases of oxygen (9%) and nitrogen (91%) at 300, 450, 600 °C, and in nitrogen at 750 and 900 °C only. The final pyrolysis vapor was directly transferred to the GC/MS and analyzed.

The chromatographic separation was performed using a DB-5MS fused silica capillary column (60 m × 250 μm id × 0.25 μm df, Agilent). The injector temperature was kept at 300 °C. Initial oven temperature was set at 50 °C, then heated to 80 °C at the rate of 6 °C min^{-1}, followed by a heating rate of 4 °C min^{-1} to 110 °C, held there for 2 min and finished at 250 °C with a rate of 5 °C min^{-1}. Helium at a constant flow rate of 1 mL min^{-1} was used as the carrier gas, and the split ratio was 100:1. The separated compounds were analyzed by the mass spectrometer. The EI ionization energy was 70 eV, and the transfer line temperature was 300 °C. Ion source temperature was 230 °C, and quadrupole temperature was 150 °C. The mass spectra were obtained from *m*/*z* 30 to 500, and solvent delay time was 3.9 min. The pyrolysis products were identified by comparison between the experimental mass spectrum and mass spectrum library (NIST 11) attached to the Py–GC/MS apparatus. All quantitative data were expressed by average values of the duplicate pyrolysis runs. For each product, its peak area% value obtained under different pyrolysis conditions can be compared to reveal the changing of its relative content among the detected products.

Results and discussion

Structure identification

The synthetic route of DMPOE and TMPOE is shown in Scheme 1. The chemical structures of the two samples were new and identified by techniques of ^{1}H NMR, ^{13}C NMR, IR and HRMS (see Table 1). These spectra diagrams were included in electronic supplementary material.

Thermal analysis of DMPOE and TMPOE samples

The TG curves and DTG curves of DMPOE and TMPOE during degradation at the heating rate of 10 °C min^{-1} are presented in Figs. 1 and 2, respectively. In Fig. 1, it was observed that the main stage was between 170 and 420 °C, and there was a sharp decrease in mass loss of 95.85%. The maximum decomposition rate of DMPOE was achieved at T_p of 310 °C with 60.70% mass loss from the beginning. From 420 to 480 °C, the sample exhibited a slow mass loss process with mass loss of 2.17%. Finally, DMPOE sample remained a solid residue and equaled to 1.34% of its original mass.

Figure 2 shows that the mass of TMPOE sample decreased with the rise of temperature. It revealed that the main degradation stage was lost about 98.37% of the total mass between 110 and 270 °C. In this stage, mass loss rate was largest appeared at T_p of 250 °C and mass loss was about 81.08% from the beginning. The last stage was high temperature charring of the residue, and the mass loss was about 0.64% of original mass. Finally, the mass of TMPOE remained 0.10% of its original mass.

TG analysis indicated that the DMPOE and TMPOE sample had good stability at room temperature, and the breaking temperature of DMPOE was higher than that of TMPOE. These phenomena might be attributed to the difference on physical form and chemical structure of DMPOE and TMPOE, which resulted in different pyrolysis reactions.

The DSC curves of DMPOE and TMPOE are shown in Figs. 3 and 4, respectively. The enthalpy change of the samples and the peak temperature of DSC curves were recorded by apparatus. It presented that the T_peak of DMPOE and TMPOE in DSC curves was 301.8 and 260.0 °C, respectively. The onset temperature of thermal decomposition was measured to help us determining the thermal stability of samples from another point. In Fig. 3, melting point of DMPOE measured by DSC and TG was 87.6 °C. T_onset, T_end and ΔH of the samples in DSC curves could be detected by apparatus. In main mass loss regions of samples, DSC analysis of DMPOE and TMPOE both showed the endothermic decomposition. In general, the results illustrated in TG–DTG and DSC curves were corresponding to each other. The data concerning the decomposition temperatures are summarized in Table 2.

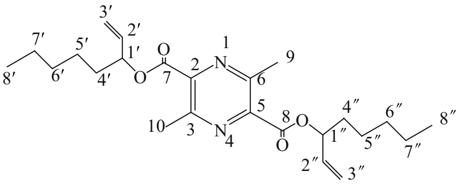
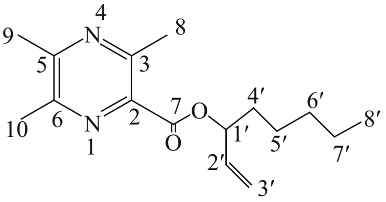
Py–GC/MS analysis of DMPOE and TMPOE samples

Although TG and DSC seemed to be significant means to study thermal properties of organic compounds, the pyrolysis products were not identified. From results of the TG and DSC analysis, the samples were pyrolyzed at 300 °C. These TG and DSC results were the verification for the pyrolysis temperatures. Py–GC/MS was the useful means to indicate the products distribution under different temperatures. In order to further understand the influence of chemical structure and pyrolysis conditions on the samples decomposition process, Py–GC/MS experiments were conducted based on the composition detection of pyrolysis products from DMPOE and TMPOE.

The research on the pyrolysis products was important for the application of DMPOE and TMPOE in tobacco. The pyrolysis conditions were in atmosphere of nitrogen and 9% oxygen in nitrogen at 300, 450 and 600 °C, and just in nitrogen at 750 and 900 °C.

Figure 5 shows Py–GC/MS detection of products evolved from pyrolysis of DMPOE and TMPOE under the pyrolysis conditions. Tables 3 and 4 list the main components released from the two samples. As shown in Table 3, the main compound types evolved out from the pyrolysis of DMPOE were diverse and the relative contents also showed great differences. As far as the pyrolysis of TMPOE was concerned, shown in Table 4, no obvious difference was observed for the pyrolysis product type in atmosphere of N_2 and 9% O_2 in nitrogen at 300, 450 and 600 °C, whereas more pyrolysis compound types were detected at 750 and 900 °C. By analysis on the volatile pyrolysis compounds of DMPOE and TMPOE, it could be found that 1-octen-3-ol and 2,5-dimethyl-3-propylpyrazine were the common products. 1,3-Octadiene, 2,4-octadiene and (*E*)-2-octen-1-ol were the conversion products of 1-octen-3-ol, the relative yields of them varied greatly. 1,3-Octadiene and 2,4-octadiene were isomer formed from the

Table 1 Characterization of the two synthesized compounds

Compounds	^{1}H NMR δ/ppm	^{13}C NMR δ/ppm	IR/cm^{-1}	HRMS
 DMPOE	0.89(t, *J* = 6.88 Hz, 6*H*, 3*H*-8′, 3*H*-8″), 1.26–1.37(m, 8*H*, 2*H*-5″, 2*H*-6″, 2*H*-6′, 2*H*-5′), 1.38–1.47(m, 4*H*, 2*H*-7′, 2*H*-7″), 1.69–1.78(m, 2*H*, H-4′, H-4″), 1.80–1.89(m, 2*H*, H-4′, H-4″), 2.75(s, 6*H*, 3*H*-9, 3*H*-10), 5.27(d, *J* = 10.44 Hz, 2*H*, H-3′, H-3″), 5.40(d, *J* = 17.24 Hz, 2*H*, H-3′, H-3″), 5.54(q, *J* = 6.72 Hz, 2*H*, H-1′, H-1″), 5.87–5.95(m, 2*H*, H-2′, H-2″)	13.96(C-8′, C-8″), 22.21(C-9, C-10), 22.49(C-7′, C-7″), 24.75(C-5′, C-5″), 31.48(C-6′, C-6″, C-4′, C-4″), 76.70(C-1′, C-1″), 118.07(C-3′, C-3″), 135.78(C-2′, C-2″), 144.49(C-5, C-2), 150.25(C-3, C-6), 164.73(C-7, C-8)	2935($\nu_{\text{C–H}}$), 2872($\nu_{\text{C–H}}$), 1720($\nu_{\text{O=C}}$), 1262($\nu_{\text{C–O–C}}$), 1127($\nu_{\text{C–O–C}}$), 987($\delta_{\text{=C–H}}$), 940($\delta_{\text{=C–H}}$)	$[M + H]^+$ 417.2740 (calcd. 417.2753), $[M + Na]^+$ 439.2578 (calcd. 439.2573)
 TMPOE	0.89(t, *J* = 6.96 Hz, 3*H*, 3*H*-8′), 1.26–1.37(m, 4*H*, 2*H*-5′, 2*H*-6′), 1.38–1.46(m, 2*H*, 2*H*-7′), 1.69–1.78(m, 1*H*, H-4′), 1.80–1.88(m, 1*H*, H-4′), 2.55(s, 6*H*, 3*H*-8, 3*H*-10), 2.71(s, 3*H*, 3*H*-9), 5.25(dt, *J* = 10.48, 1.0 Hz, 1*H*, H-3′), 5.39(dt, *J* = 17.24, 1.16 Hz, 1*H*, H-3′), 5.53(q, *J* = 6.76 Hz, 1*H*, H-1′), 5.88–5.96(m, 1*H*, H-2′)	13.95(C-8′), 21.57(C-8, C-10), 22.10(C-9), 22.47(C-7′), 24.77(C-5′), 31.50(C-6′), 34.09(C-4′), 76.73(C-1′), 117.67(C-3′), 136.12(C-2′), 140.36(C-2), 149.36(C-6), 150.39(C-5), 153.96(C-3), 165.55(C-7)	2932($\nu_{\text{C–H}}$), 2861($\nu_{\text{C–H}}$), 1722($\nu_{\text{O=C}}$), 1299($\nu_{\text{C–O–C}}$), 1174($\nu_{\text{C–O–C}}$), 987($\delta_{\text{=C–H}}$), 931($\delta_{\text{=C–H}}$).	$[M + H]^+$ 277.1934 (calcd. 277.1916), $[M + Na]^+$ 299.1732 (calcd. 299.1735)

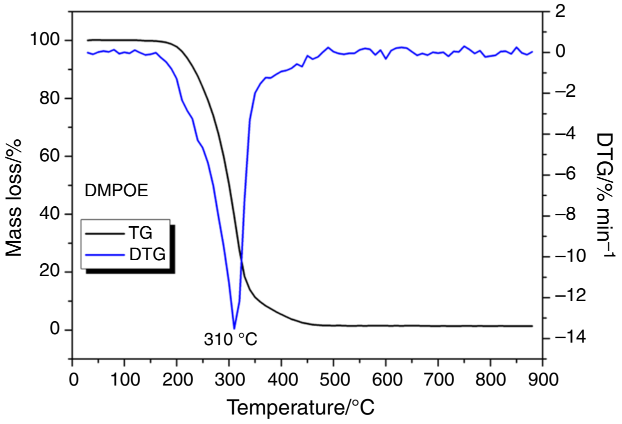


Fig. 1 TG and DTG curves of DMPOE

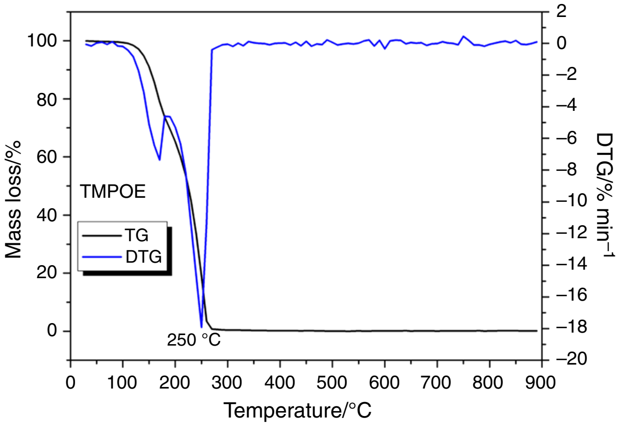


Fig. 2 TG and DTG curves of TMPOE

elimination reaction of 1-octen-3-ol, while 2,4-octadiene could be generated from 450 to 900 °C with little amount. The result of rearrangement of 1-octen-3-ol under high heating temperature gave rise to formation of (*E*)-2-octen-1-ol, which could continuously underwent the loss of water to generate 1,3-octadiene. The other main alkylpyrazines resulting from DMPOE pyrolysis in Table 3 are 2,5-dimethylpyrazine, 2-ethyl-5-methylpyrazine, 2-isobutyl-3-methylpyrazine and 2-methyl-5-propylpyrazine. As shown in Table 4, the other main alkylpyrazines resulting from

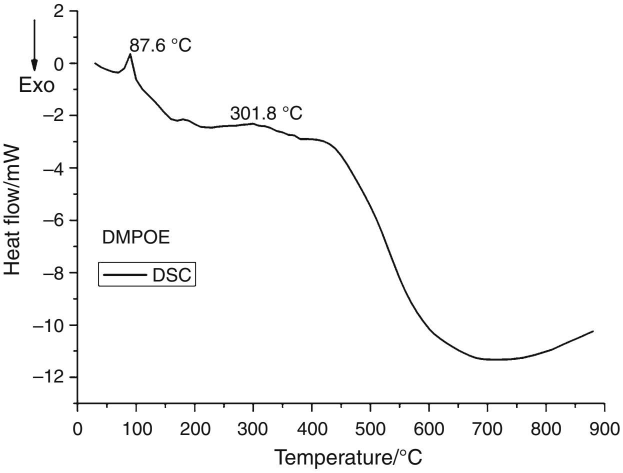


Fig. 3 DSC curve of DMPOE

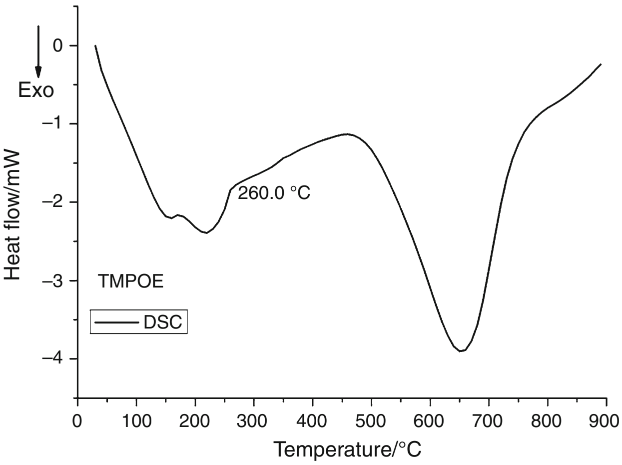


Fig. 4 DSC curve of TMPOE

TMPOE pyrolysis are 2,6-dimethylpyrazine, 2,3,5-trimethylpyrazine, 3-ethyl-2,5-dimethylpyrazine, 2-ethyl-3,5-dimethylpyrazine, 2,3-dimethyl-5-ethylpyrazine and 2,5-dimethyl-3-pentylpyrazine. Among all pyrolysis products of the two samples, 1-octen-3-ol, 2,3,5-trimethylpyrazine, 2,5-dimethylpyrazine, 2-ethyl-3,5-dimethylpyrazine, 2-isobutyl-3-methylpyrazine, 2,3-dimethyl-5-ethylpyrazine and 2,5-dimethyl-3-propylpyrazine were the characteristic flavor compounds we expected. Some of them could be detected in Cocoa products [12]. Meanwhile, Adams et al. [31] reported that the FEMA number of 2,3,5-trimethylpyrazine was 3244 and 2,5-dimethylpyrazine was 3272. These alkylpyrazines were thermal degradation compounds or rearrangement compounds during high-temperature conditions.

Table 3 shows that 1,3-octadiene, 2,5-dimethylpyrazine, 1-octen-3-ol and 2-isobutyl-3-methylpyrazine were generated by DMPOE degradation under every pyrolysis condition. 1,3-Octadiene, 2,4-octadiene and (*E*)-2-octen-1-ol were detected in the volatiles, among which 1,3-octadiene accounted for a large proportion. In Table 3, the relative amount of the characteristic flavor compound of 2,5-dimethylpyrazine was increased with the rising of temperature. The maximum amount of 1-octen-3-ol was 12.30% at 600 °C in N_2 atmosphere. 2-Isobutyl-3-methylpyrazine existed in coffee [12], amount of which achieved maximum amount of 20.35% at 750 °C. Flavor compound of 2,5-dimethyl-3-propylpyrazine was produced only at 300 °C with relative content of 44.61% and 57.35% in two atmosphere and might transform into 2-methyl-5-propyl pyrazine at the higher temperature.

From Table 4, it was observed that 1,3-octadiene, 2,3,5-trimethylpyrazine, 2,5-dimethyl-3-propylpyrazine and 2,5-dimethyl-3-pentylpyrazine were produced from TMPOE under every pyrolysis condition. Although 1,3-octadiene, 2,4-octadiene and (*E*)-2-octen-1-ol were also detected, their relative contents were comparatively lower than that of DMPOE pyrolysis. This was due to the proportion of 1-octen-3-ol existed in their original structure of the samples. The amount of 2,3,5-trimethylpyrazine attained the maximum amount of 23.03% at 750 °C. 2,5-Dimethyl-3-propylpyrazine accounted for the most proportion compared to other products on every pyrolysis condition; nevertheless, the relative content of 2,5-dimethyl-3-pentylpyrazine was lower than that of the former. It was considered that 2,5-dimethyl-3-propylpyrazine was the main pyrolysis product and formed easily in all pyrolysis products.

It was observed that DMPOE and TMPOE could be used as flavor precursors to release characteristic flavors under similar cigarette combustion conditions. Meanwhile, the Py–GC/MS analysis indicated that the sample chemical structure had obvious significance on the type and relative amount of pyrolysis products. Combining TG with DSC analysis, it was the cleavage of ester bond of samples to release the pyrolysis products and rearranged to produce other useful flavor compounds.

Table 2 Thermal analysis data for DMPOE and TMPOE

Sample name	DSC				TG–DTG	
	T_{onset}/°C	T_{peak}/°C	T_{end}/°C	ΔH/kJ mol^{-1}	T_p/°C	T_{range}/°C
DMPOE	240.4	301.8	359.4	62.44	310	170–420
TMPOE	247.9	260.0	272.1	25.84	250	110–270

Fig. 5 Typical ion chromatograms from pyrolysis of the DMPOE and TMPOE at different temperatures and atmospheres (peak identification referred in Tables 3 and 4, respectively). Pyrolysis conditions were set at 300, 450 and 600 °C in atmosphere of N_2 and mixed gases of 9% oxygen and 91% nitrogen, as well as at 750 and 900 °C in N_2 only

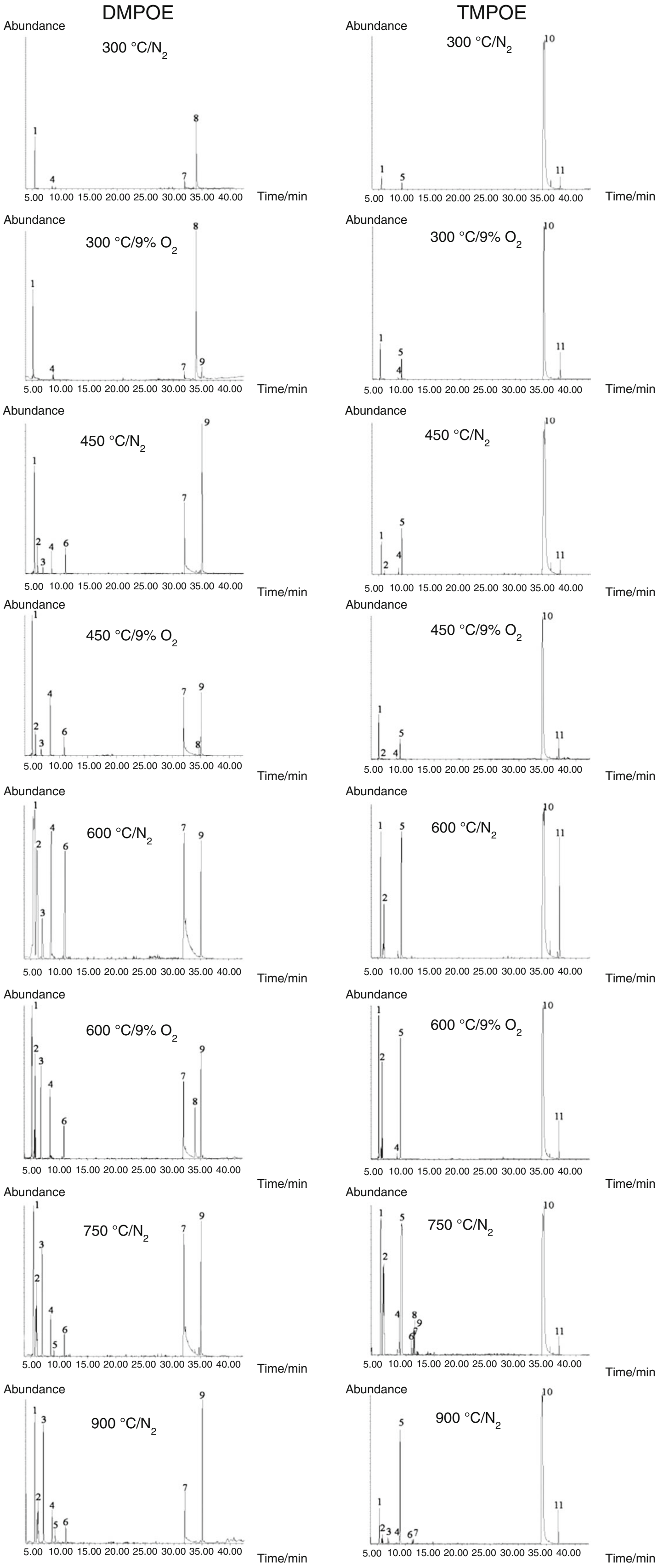
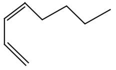
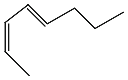
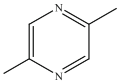
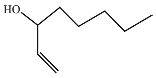
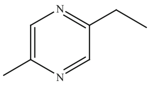
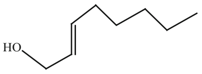
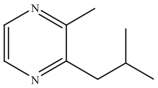
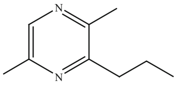
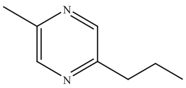


Table 3 Pyrolysis products of DMPOE

Peak no.	RT /min	Compounds	300 °C		450 °C		600 °C		750 °C	900 °C
			N_2	9% O_2	N_2	9% O_2	N_2	9% O_2	N_2	N_2
1	5.43	 1,3-Octadiene	47.06	25.33	20.89	36.83	30.83	26.74	23.26	23.77
2	5.98	 2,4-Octadiene	–	–	3.65	2.82	15.65	9.70	10.79	9.78
3	6.92	 2,5-Dimethylpyrazine	0.46	0.54	0.92	1.60	4.69	7.52	11.80	18.35
4	8.47	 1-Octen-3-ol	0.98	1.71	2.79	10.04	12.30	6.75	3.00	3.56
5	9.05	 2-Ethyl-5-methylpyrazine	–	–	–	–	–	–	0.59	1.73
6	10.89	 (E)-2-Octen-1-ol	–	–	3.39	3.73	7.76	3.05	1.46	1.66
7	31.92	 2-Isobutyl-3-methylpyrazine	2.26	2.90	13.65	18.83	13.88	13.16	20.35	6.11
8	33.99	 2,5-Dimethyl-3-propylpyrazine	44.61	57.30	–	0.82	–	8.43	–	–
9	34.73	 2-Methyl-5-propylpyrazine	–	1.02	44.81	13.87	5.34	16.06	17.66	22.96

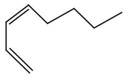
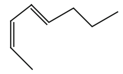
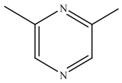
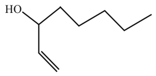
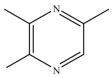
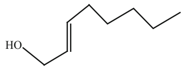
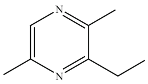
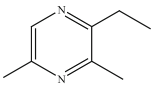
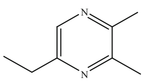
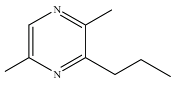
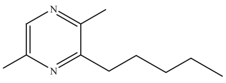
RT retention time, – compound was undetected

N_2 means the pyrolysis atmosphere was only in nitrogren. 9% O_2 means that the atmosphere was in the mixed gases of 9% oxygen and 91% nitrogen

Pyrolysis mechanism analysis

The study on the formation of characteristic pyrolysis products was considerable for potential application of DMPOE and TMPOE as flavor additives in tobacco. Under the cigarette burning conditions, the possible pyrolysis pathways of the two samples were proposed [32–34]. The pyrolysis products were generated ultimately resulting from the interaction among pyrolysis temperature and atmosphere. From Tables 3 and 4, the pyrolysis temperatures were influential, while the influence of pyrolysis atmospheres on the types and contents of pyrolysis

Table 4 Pyrolysis products of TMPOE

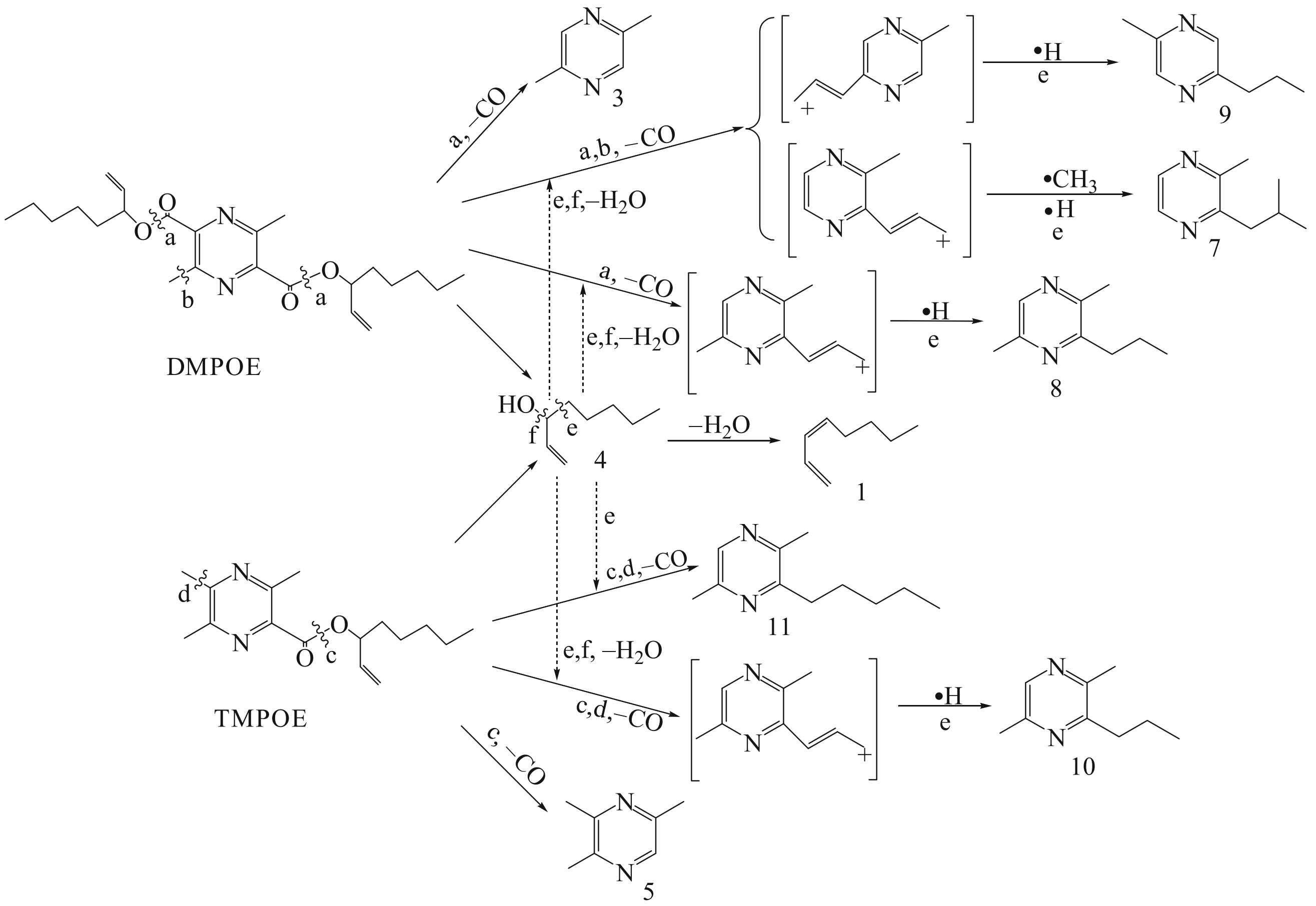
Peak no.	RT /min	Compounds	300 °C		450 °C		600 °C		750 °C	900 °C
			N_2	9% O_2	N_2	9% O_2	N_2	9% O_2	N_2	N_2
1	5.14	 1,3-Octadiene	1.78	4.64	3.69	5.19	10.93	11.63	15.49	3.26
2	5.98	 2,4-Octadiene	–	–	0.27	0.08	5.22	5.19	10.25	1.20
3	7.01	 2,6-Dimethylpyrazine	–	–	–	–	–	–	0.73	0.54
4	8.33	 1-Octen-3-ol	–	0.27	0.38	0.19	0.51	0.28	1.21	0.13
5	8.94	 2,3,5-Trimethylpyrazine	0.57	2.36	2.90	2.05	11.51	12.72	23.03	10.98
6	10.95	 (E)-2-Octen-1-ol	–	–	–	–	–	–	0.33	0.20
7	11.27	 3-Ethyl-2,5-dimethylpyrazine	–	–	–	–	–	–	0.70	0.25
8	11.43	 2-Ethyl-3,5-dimethylpyrazine	–	–	–	–	–	–	0.71	0.24
9	11.50	 2,3-Dimethyl-5-ethylpyrazine	–	–	–	–	–	–	0.82	0.29
10	34.30	 2,5-Dimethyl-3-propylpyrazine	95.61	86.69	89.06	86.87	60.53	67.13	42.32	78.18
11	37.06	 2,5-Dimethyl-3-pentylpyrazine	1.02	3.43	0.87	2.35	7.67	2.45	0.55	2.32

RT retention time, – compound was undetected

N_2 means the pyrolysis atmosphere was only in nitrogren. 9% O_2 means that the atmosphere was in the mixed gases of 9% oxygen and 91% nitrogen products was not obvious. As shown in Scheme 2, the 1,3-octadiene (peak no. **1**) was formed from dehydration of 1-octen-3-ol (peak no. **4**) in both atmosphere of N_2 and 9% O_2 in nitrogen. High heating temperature was beneficial to the formation of various radicals, which could promote the formation of pyrolysis products.

From the pyrolysis products of DMPOE and TMPOE, it was believed that the cleavage of ester bond of C-O could be carried out in the decomposition process. As shown in Scheme 2, for DMPOE and TMPOE, the chemical bond **a** and **c** ruptured and then left CO to produce the aroma compounds of 2,5-dimethylpyrazine with peak no. **3** and 2,3,5-trimethylpyrazine with peak no. **5**, respectively. At the moment, 1-octen-3-ol (peak no. **4**) was obtained and could easily undergo elimination reaction to form 1,3-octadiene (peak no. **1**). The bonds **e** and **f** of 1-octen-3-ol broke simultaneously, and H_2O molecule was eliminated to generate allyl carbocation and pentyl radical. For DMPOE, the chemical bond **a** and **b** broke and then eliminated CO. After that, allyl carbocation was captured in *para*-position of 2-methylpyrazine to produce 2-methyl-5-propylpyrazine (peak no. **9**) with the capture of three H radicals and one electron. Meanwhile, allyl carbocation was captured in *ortho*-position of 2-methylpyrazine and then underwent addition reaction with CH_3 and H radicals to afford 2-isobutyl-3-methylpyrazine (peak no. **7**) with the capture of an electron. With breakage of the bond **a** and elimination of CO, 2,5-dimethyl-3-propylpyrazine (peak no. **8**) was produced through the capture of allyl carbocation, three H radicals and one electron.

For TMPOE, the pyrolysis process was similar to that of DMPOE. The bond **c** and **d** broke and then eliminated CO. Afterward, 2,5-dimethyl-3-pentylpyrazine (peak no. **11**) was formed by capturing one pentyl radical. Meanwhile, allyl carbocation was captured and then three H radicals were added. These reactions resulted in the generation of 2,5-dimethyl-3-propylpyrazine (peak no. **10**) with the capture of one electron. In addition to the above pyrolysis procedures of the samples, some radicals reacted or rearranged to produce other compounds [35].



Scheme 2 Proposed pyrolysis pathways for DMPOE and TMPOE. The *numbers* represented the peak number of identified compounds

Conclusions

The flavor precursors of DMPOE and TMPOE were synthesized, and their structures were confirmed by ^{1}H NMR, ^{13}C NMR, IR and HRMS. The thermal behavior and decomposition compounds of DMPOE and TMPOE were discussed by TG, DSC and Py–GC/MS analysis. TG analysis indicated that the DMPOE and TMPOE had good stability at room temperature, whereas the breaking temperature of DMPOE was higher than that of TMPOE. T_p of DMPOE and TMPOE was 310 and 250 °C, respectively. DSC curves demonstrated that T_peak was 301.8 °C for DMPAOE and 260.0 °C for TMPAOE with endothermic decomposition. Py–GC/MS analysis showed that there were some characteristic aroma compounds produced such as 1-octen-3-ol, 2,3,5-trimethylpyrazine, 2,5-dimethylpyrazine, 2-isobutyl-3-methylpyrazine and 2,5-dimethyl-3-propylpyrazine. The sample chemical structure had obvious significance on the type and relative amount of pyrolysis products. The influence of pyrolysis atmospheres was not obvious. The pyrolysis mechanisms of DMPOE and TMPOE were proposed. Under pyrolysis condition, the primary decomposition reaction was that DMPOE and TMPOE broke down to regenerate the corresponding characteristic flavor products.

Supporting information

The ^{1}H NMR, ^{13}C NMR, IR and HRMS spectra diagrams of oct-1-en-3-yl methylpyrazinecarboxylates are available to authorized users.

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