

## Analysis of Volatile Flavour Compounds in Unloosed and Loosed Tobaccos

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The volatile flavour compounds of tobaccos were analyzed by capillary gas chromatography (GC). Volatile flavour compounds were isolated by dichloromethane extraction followed by steam distillation continuous extraction (SDE). In total, 41 volatiles were identified. They consisted mainly of compounds exhibiting aromatic characteristics. The quality and quantity of volatile flavour compounds exhibited different distribution in unloosed or loosed tobaccos. Compared with unloosed tobaccos, volatile flavour compounds showed a decrease after the loosing process.

**Key Words:** Tobacco, Volatile flavour compounds, Loosing Process, Capillary GC.

### INTRODUCTION

At present, with the assistance of advanced analyzer technology, many analysts of tobacco research organizations or cigarette enterprises are devoting themselves to reveal the rule of changes of chemical components during cigarette primary processing, confirm the interrelation between the changes of main chemical components in tobacco and the processing parameters<sup>1, 2</sup>. The evaluation of the quality of cigarette process through organoleptic evaluation combined with chemical evaluation comes true step by step. Various theories provided a base to improve the quality of tobacco processing, only tobacco casing and dryer<sup>3-5</sup>.

The process of loose tobacco involves the first heating and ordering process with a little attribute for tobacco processing; the changes of chemical components have not been noticed by the researchers.

In this investigation, steam distillation continuous extraction (SDE) and capillary gas chromatography (GC) were used to analyze the changes of volatile flavour components during loosing tobacco in cigarette processing. 41 Flavor components were identified. During loosing tobacco, the changes of volatile flavour components were distinct.

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## EXPERIMENTAL

Loosed and unloosed flue-cured tobaccos ("SEPTWOLVES<sup>®</sup>", Longyan Cigarette Manufactory of China) were collected from processing lines. 1 kg of each sample was collected and mixed thoroughly, put into 40°C oven for 8 h to remove moisture, then ground to 40–60 mesh powder.

All solvents employed (including anhydrous dichloromethane, anhydrous ethanol and heptadecane) were of analytical grade quality and redistilled before use. Standards of volatile flavour compounds were all purchased from Sigma (St. Louis, MO) and were all in GC purities. Standard solutions were used to optimize GC/MS and GC conditions. All were refrigerated at 4°C during storage.

### Extraction of volatile flavour compounds

25.0 g ground tobacco (40–60 meshes) in 250 mL flask, added with 350 mL distilled water and a little zeolite. By use of a Likens-Nickerson steam distillation continuous extraction head<sup>6</sup> with 50 mL dichloromethane as extraction solvent, the mixture was extracted and refluxed for 2.5 h at atmospheric pressure. Different volatile flavour compounds were separated as the step shown in Fig. 1. 5.0 µL methyl-nicotinic acid was added as an internal standard to indicate to each separate fraction which was dried over anhydrous sodium sulfate. The diethyl ether extract was transferred into a K.D. concentrator and concentrated to 1 µL was used for GC or GC/MS analysis.

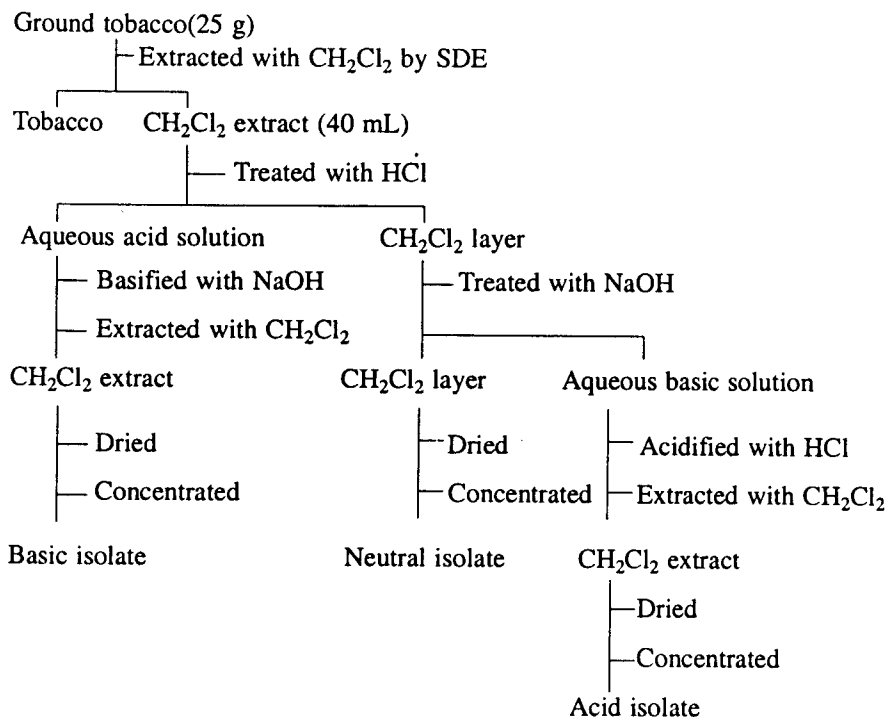


Fig. 1. Separation steps for the acidic, basic and neutral volatile flavour compounds.

### Capillary GC analysis

(1) **GC analysis of acid volatile flavour compounds:** FFAP capillary chromatographic column (50 m × 0.2 mm id × 0.33 μm d.f.); the flow velocity of carrier gas (N<sub>2</sub>): 1.0 mL/min; injector: 270°C; FID: 270°C; split ratio: 15 : 1; temp. ramp: 70°C (1 min)—4°C/min—210°C.

(2) **GC analysis of alkaline volatile flavour compounds:** ULTRA-2 capillary chromatographic column (50 m × 0.25 mm id × 0.33 μm d.f.); detector: NPD; H<sub>2</sub> flow: 2.0 mL/min; air flow: 60 mL/min; carrier gas (He) flow: 1.0 mL/min; injector: 250°C; split ratio: 10 : 1; temp ramp: 50°C (1 min)—2°C/min—200°C—4°C/min—260°C(5 min).

(3) **GC analysis of neutral volatile flavour compounds:** ULTRA-2 capillary chromatographic column (50 m × 0.25 mm id × 0.33 μm d.f.); detector: FID; H<sub>2</sub> flow: 40 mL/min; air flow: 400 mL/min; carrier gas (He) flow: 1.0 mL/min; injector: 250°C; split ratio: 10 : 1; temp. ramp: 50°C (1 min)—2°C/min—200°C—4°C/min—260°C (5 min).

### Qualitative and quantitative analysis

Qualitative analysis was carried out directly by the retention time of the standard compounds, use methyl-nicotinic acid as an internal standard, quantitative analysis was carried out by peak area by calculating quantitative calibrate factor for each according to chromatographic peak area of acid, alkaline and neutral flavour compounds.

## RESULTS AND DISCUSSION

### Analyses the changes of neutral flavour compounds

15 Kinds of neutral volatile flavour compounds were identified. In loosed tobaccos, the neutral volatile flavour components decreased by 7.48% (Table-1). The contents of benzaldehyde increased by 28.91% and β-phenylethyl alcohol increased by 17.92%. Study of tobacco and cigarette showed that properly increasing of benzaldehyde and β-phenylethyl alcohol could gives contributes in richness, body and some delicate taste to cigarette smoke<sup>7-12</sup>.

### Analyses the changes of basic flavour components

14 Kinds of neutral volatile flavour components were identified. In loosed tobaccos, the alkaline volatile flavour components decreased by 13.38%. Pyrrole, 2,3-diethylpyrazine and indol decreased significantly, by 96.12, 56.52 and 47.67% respectively (Table-1). Study of tobacco and cigarette showed that decreasing of alkaline properties could reduce the harshness, irritation to mouth and nose and the impact on throat<sup>7-12</sup>.

TABLE-1  
 QUANTITATIVE RESULTS OF NEUTRAL, BASIC, ACIDIC FLAVOUR COMPOUNDS  
 IN UNLOOSED AND LOOSED TOBACCOS ( $\mu\text{g g}^{-1}$ )

Peak No.	Retention (min)	Compounds	Pre loosening	Post loosening
Neutral 1.	11.07	2-Methyl-tetrahydrofuran-3-one	44.78	33.70
2.	12.40	2-Furfuraldehyde	298.56	230.57
3.	13.50	Furanmethanol	108.33	90.49
4.	22.04	6-Methyl-5-hepten-2-one	3.94	3.52
5.	25.43	Benzaldehyde	101.92	131.39
6.	25.77	2-Acetyl-5-methylfuran	6.36	trace
7.	30.16	Linalool	10.36	9.80
8.	31.26	$\beta$ -phenylethyl alcohol	48.95	57.72
9.	31.78	Isophorone	3.02	3.27
10.	33.44	Isophorone-oxide	36.66	36.81
11.	39.52	Citronellol	46.36	46.53
12.	49.98	$\beta$ -damascone	48.15	48.32
13.	51.88	$\beta$ -damascenone	52.28	52.49
14.	56.36	$\beta$ -ionone	45.03	46.48
15.	59.22	5,6,7,7a-Tetrahydro-2(4H)-benzofuranone	2.77	2.23
	Total		857.47	793.32
Basic 1.	9.21	Thiazole	0.11	0.08
2.	9.66	Pyridine	4.72	3.48
3.	9.84	Pyrrole	1.03	0.04
4.	18.40	2,5-Dimethylpyrazine	0.02	0.02
5.	19.00	2,3-Dimethylpyrazine	0.46	0.20
6.	24.44	2,3,5-Trimethylpyrazine 2,3,5-Trimethylpyrazine	1.28	1.10
7.	24.80	2-3-Dimethylpyrazine	2.16	2.03
8.	27.00	2-Acetylpyrrole	0.57	0.61
9.	28.80	2,3-Diethylpyrazine	137.31	122.00
10.	30.53	2,3,5,6-Tetramethylpyrazine	—	0.28
11.	30.78	2-Acetyl-1-Methyl-pyrrole	1.38	1.80
12.	32.32	3-Acetylpyridine	1.39	1.57
13.	41.75	Quinoline	0.51	0.51
14.	45.41	Indol	8.60	4.50
	Total		159.54	138.2
Acidic 1.	19.36	Propanoic acid	9.02	11.36
2.	20.23	Isobutyric acid	13.55	15.43
3.	22.14	Butyric acid	8.63	8.72

Peak No.	Retention (min)	Compounds	Pre loosening	Post loosening
4.	23.01	Isovaleric acid	128.32	150.70
5.	24.92	Pentanoic acid	68.32	79.74
6.	25.93	2-Methylpentanoic acid	172.48	197.56
7.	28.02	Hexanoic acid	101.74	122.65
8.	35.93	Heptanoic acid	58.31	64.27
9.	36.02	Nonanoic acid	75.67	123.30
10.	38.85	Decanoic acid	63.41	79.16
11.	45.12	Benzoic acid	209.70	224.52
12.	46.04	Laurate	138.58	192.60
	Total		1047.73	1270.01

### Analyses the changes of acidic flavour components

12 Kinds of acid volatile flavour components were identified. In loosed tobaccos, the acid volatile flavour components increased by 21.22%. The content of propanoic acid, isobutyric acid, butyric acid and heptylate acid increased a little, but the other 8 kinds of acid increased significantly (Table-1). Study of tobacco and cigarette showed that increasing of acid flavour components properties in tobacco, gives a mellowing, smoothing effect to the smoke taste and reduces irritation or harshness in mouth and nose<sup>7-12</sup>.

### Conclusion

SDE coupled with GC was used for qualitative and quantitative analysis of the changes of volatile acid, neutral, basic flavour components in unloosed and

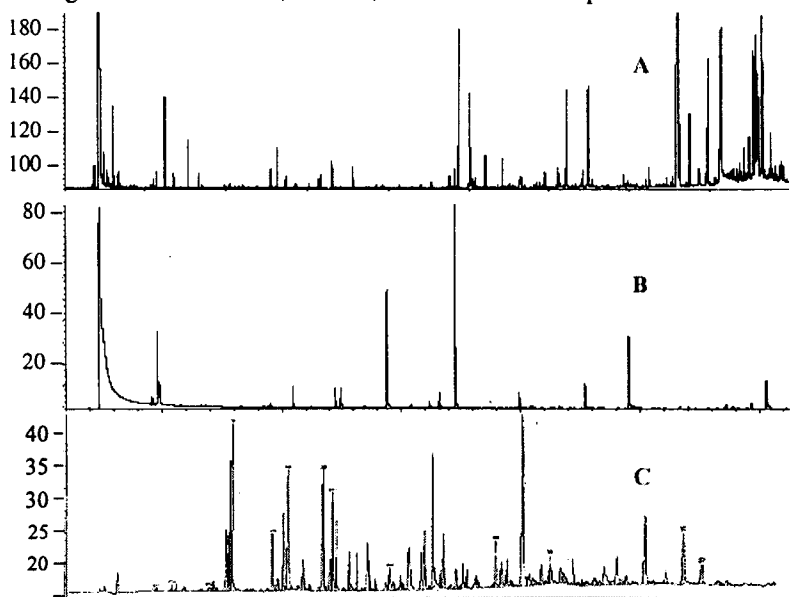


Fig. 2. Chromatogram of neutral (A), basic (B) and acidic (C) flavour compounds

loosed tobaccos of the cigarette manufacture processing. This investigation showed that during loosing tobacco, the changes of volatile flavour components were distinct. The neutral volatile flavour components decreased by 7.48%. The alkaline volatile flavour components decreased by 13.38%, while the acid volatile flavour components increased by 21.22%. The changes of volatile flavour compounds were all helpful to organoleptic quality. For tobacco industry, the conclusion of this investigation could have some useful to processing control. The chemical components of tobacco can effected the cigarette quality not only by the process such as tobacco dry at a high temperature or casing<sup>6</sup>, but also by the process such as loosing tobaccos.

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