



## Effects of Plant Volatiles on the EAG Response and Behaviour of *Saperda populnea* L. (Coleoptera: Cerambycidae)

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Essential oil in the branches of eleven varieties of *Populus L.* at different periods were extracted using the steam distillation method and the chemical composition and relative content of volatile oil from these branches were investigated by GC-MS technology. EAG response of *Saperda populnea L.* adults to the volatile oil from branches and their behavioural response in the Y-tube olfactometer were tested. The results shown that the male and female adults of *S. populnea* were more responsive and showed obvious selectivity to volatile oil from *P. aigeiros* duby section such as *P. deltoides* Marsh  $\times$  *P. cathayana* Rehd., *P. simonii* Carr.  $\times$  *P. nigra* L. and *P.  $\times$  euramericana* 'DN113' than to the volatile oil from *P. davidiana* Dode  $\times$  *P. bolleana* Lauche and *P. alba* L.  $\times$  *P. berolinensis* Dipp. The main content of essential oil were alcohols, ketones, alkanes, terpenes and heterocyclic compounds; 1,2-cyclohexanedione and 2-cyclohexen-1-one existed in the essential oil at growing and dormancy period of all of the tested poplar varieties and thus can be speculated as specific odor components or their derivatives. The relative content of aromaticity components in essential oil was higher in growing plants than that in dormant plants.

**Key Words:** EAG, GC-MS, *Saperda populnea L.*, Volatile oil, Y-tube olfactometer.

### INTRODUCTION

Plant volatiles, also known as essential oils, usually refers to a class of oily substances that have < 250 u molecular weight and < 340 °C boiling point and can be steamed out with the water vapour. Generally, they can be divided into two categories according to the nature of plant odor: 1) specific odor components, such as allyl isothiocyanate released from cruciferae plants; 2) general odor component, usually referring straight-chain alcohols, aldehydes and lipid compounds, unsaturated fatty acids, terpenoids, *etc.*<sup>1,2</sup>. That contain six carbon atoms. Plant volatiles leave the surface of plants and spread to the surrounding. Olfactory organs of herbivorous insects can gradually identify these volatile substances during coordinating evolution and thus, these volatile substances play important roles in the host identification process of pest<sup>3-5</sup>. It is reported that healthy pine volatiles can lure *Monochamus*<sup>6</sup>. This indicates that according to the species of phytophagous insects, it is easier to filter out strong moth-attractive substances from the volatile material components in leaves and plants at different seasons or different developmental stages of host plants or plants with preferred odor.

Poplar is the general name of salicaceae *Populus L.* plants and it is widely distributed in China. The total area of poplar plantation worldwide is about 800 million hm<sup>2</sup> and there is about 660 million hm<sup>2</sup> in China, ranking first in the world<sup>1,7</sup>.

But in recent years, some poplar saplings are damaged by *Saperda populnea L.* *Saperda populnea L.* belong to coleoptera *Saperda*, also known as *Saperda populnea*. It is mainly distributed in northeast, north, northwest China. The larvae eat branches, especially the shoot part. The damaged positions formed spindle-shaped galls, which impede the normal transport of nutrients, making the shoot dry and vulnerable to wind-caused break. If larvae damage backbone of the saplings, the whole plant can die. With the rapid development of chemical ecology, growing emphasis has been placed on the chemical link research between insect and plant, hoping to identify chemical ecology materials that can effectively regulate pest population<sup>8-10</sup>. For example, the mixture of monoterpenes and ethanol can effectively lure *Monochamus*<sup>11,5</sup> and *Acer negundo* Linn and *Acer mono* have significant lure effects to *Anoplophora glabripennis*<sup>12</sup>. However, few studies are available about *Saperda populnea L.* and their host plant volatiles and the pest control of *Saperda populnea L.* during practice remains on pesticide spray and artificial pruning. In order to investigate the composition of volatiles in poplar branches, materials with good lure effects to *Saperda populnea L.* were screened. In this study, we extracted and separated volatiles from branches of 11 poplars at growth and dormant stages and performed composition analysis and comparison studies. The olfactory response and olfactory choice behaviour of male and female

*Saperda populnea* L. adults to these volatiles were measured by antennae potential instrument and Y-shaped olfactometer in order to filter out the effective attractant to *Saperda populnea* L.

## EXPERIMENTAL

*Saperda populnea* L. were collected from *Saperda populnea* L. damaged poplars in January 2008 at Hongqi Forest Farm, Daqing city, Heilongjiang Province, China. Galls on poplar saplings were cut at 8-10 cm away the gall, brought back to the laboratory and inserted into the moist sand in insectarium with a temperature of 24-26 °C and a relative humidity of 70-80 %. Water was regularly sprayed to the soil to maintain the moisture and energy of branches.

Eleven types of poplar branches with 3 years of age were collected in June 2009 and November 2009 from Forest and Environmental Sciences Research Institute, Qiqihar city, Heilongjiang province, China. They were labeled as A-K: A, *P. pseudo-simonii* Kitag. × *P. nigra* L.; B, *P. nigra* L.; C, *P. stalinetz* Jabl. D, *P. deltooides* Marsh × *P. simonii* Carr.; E, *P. simonii* Carr. × *P. nigra* L.; F, *P. deltooides* Marsh × *P. cathayana* Rehd.; G, *P. deltooides* Marsh × *P. suaveolens* Fisch.; H, *P. × euramericana* 'DN113'; I, *P. alba* L. × *P. berolinensis* Dipp.; J, *P. pseudo-cathayana* × *P. deltooides* Barry 'Shan Hai Guan'; K, *P. davidiana* Dode × *P. bolleana* Lauche.

**Extraction of volatile substances:** 100 g poplar saw dust was weighed from each type of poplar and 500 mL distilled water was added for distillation until distillate was absent. Distillate was extracted with 120 mL dichloromethane, 30 mL each time, four times in total. Extract was supplemented with anhydrous sodium sulfate, placed in the refrigerator for 24 h to dry and then filtrated. The filtrate was concentrated at 40 °C water bath by rotary evaporator to obtain the 11 poplar branch volatile oils with light yellow-green colour and pungent smell for GC-MS analysis.

**Gas chromatography-mass spectrometry analysis condition and mass spectra analysis:** HP6890/5973 gas chromatograph-mass spectrometer and capillary column (25 m × 200 μm × 0.33 μm) from Agilent, U.S. were used. Temperature increased from 60 °C at 3 °C/min to 200 °C (keep 10 min), using helium as carrier gas, 1 mL/min flow rate and 1 μL injection volume. Ionization: EI. electron energy: 70 eV. Ion source temperature: 230 °C. Scan range: 30-600 amu<sup>9,13,14</sup>. After GC-MS analysis, the compounds obtained from the spectra were recognized mainly through standard computer mass spectra library (NIST98) supplemented by manual identification. The relative content of each component was expressed as the percentage of mass ion peak area.

***Saperda populnea* L. olfactory choice behaviour measurement:** The measurement used self-made Y type olfactometer, with an ordinary filter paper in 4 cm<sup>2</sup> area as carrier for the odor source, 20 μL of volatile oil on the filter paper as odor source and clean filter paper as control. Each group examined 20 insects to observe the choice behavioural response of adult. Entrance into an arm within 1 min indicated tropism to this side. Entrance into the arm connected with the odor source was recorded that the insect exhibited tropism or positive tropism to the odor source; entrance into the arm connected

with the control filter paper was recorded that *Saperda populnea* L. exhibited negative tropism to the odor source; no entrance to either arm was recorded no response. Each volatile oil test was repeated five times.

**EAG response of *Saperda populnea* L.:** Antennae potentiostat (Syntech Company, Netherlands) was mainly composed by micro-manipulator INR-5, data acquisition controller IADAC-4, odor stimulus control CS-55 and antennae potential record and output device. Stimulation flow velocity and continuous flow were both 400 mL/min, stimulation time was 0.5s and the two stimulus interval was 30-60s. Qualitative filter paper was cut into 2 cm × 0.5 cm, folded into 'Z' shape and used as carrier for each solution. Each time, 10 μL sample was evenly dropped on the filter paper and placed in Pasteur tube. Antennae of *Saperda populnea* L. was cut off by sharp blade from the base, with 2-3 mm tip removed and the two ends of antennae was fixed on gel electrode by conductive adhesive. Odor mixing tube and the antenna were 1 cm away. Dichloromethane was used as control. Each sample contained three antennae repeats.

**Data analysis:** Repellent rate, lure rate and response rate were calculated by the following formula<sup>10,15</sup>:

$$\text{Repellent ratio (\%)} = \frac{\text{total insect number in control arm}}{\text{total insect number in test}} \times 100$$

$$\text{Lure rate (\%)} = \frac{\text{total insect number in treatment arm}}{\text{total insect number in test}} \times 100$$

$$\text{Response rate (\%)} = \frac{(\text{total insect number in control arm} + \text{total insect number in treatment arm})}{\text{total insect number in test}} \times 100$$

The olfactory response difference to different volatile oils in male and female *Saperda populnea* L. adults was analyzed by Tukey's multiple comparison using SPSS 17.0 software. The olfactory response difference to same volatile oil in male and female *Saperda populnea* L. adults was analyzed by two-sample T-test. The significance of EAG response value was analyzed by variance test and significance of choice behaviour response data was analyzed by paired T test.

## RESULTS AND DISCUSSION

**EAG responses of male and female *Saperda populnea* L. adults to volatile oils from branches of different poplar at growth stages:** The EAG responses of male and female *Saperda populnea* L. adults to volatile oils from branches of 11 poplars at growth stages were shown in Table-1. The absolute EAG value showed that the highest EAG response was to *P. pseudo-simonii* Kitag. × *P. nigra* L. in females and to *P. deltooides* Marsh × *P. cathayana* Rehd. in males. The absolute EAG value to different volatile oils was higher in females than in males. Tukey's multiple comparison revealed that in females, the EAG responses to *P. pseudo-simonii* Kitag. × *P. nigra* L., *P. nigra* L., *P. stalinetz* Jabl., *P. deltooides* Marsh × *P. simonii* Carr., *P. simonii* Carr. × *P. nigra* L., *P. deltooides* Marsh × *P. cathayana* Rehd. and *P. deltooides* Marsh × *P. suaveolens* Fisch. volatile oils were extremely significantly different from the EAG response to dichloromethane ( $P < 0.01$ ); the EAG response to *P. × euramericana* 'DN113' volatile oil was significantly different compared with the EAG response to dichloromethane ( $P < 0.05$ ). In males, the EAG responses to

TABLE-1  
EAG RESPONSE OF FEMALE AND MALE *Saperda populnea* L. ADULTS TO DIFFERENT ESSENTIAL OIL

Volatiles	Female EAG value	Tukey Tests	Male EAG value	Tukey tests	Two-sample T test
<i>P. pseudo-simonii</i> Kitag. × <i>P. nigra</i> L.	1.70 ± 0.25	Aa	0.55 ± 0.23	ABCabcd	0.010
<i>P. nigra</i> L.	1.58 ± 0.20	Aab	0.63 ± 0.17	ABCabc	0.008
<i>P. stalinetz</i> Jabl.	1.37 ± 0.38	Aab	0.43 ± 0.27	ABCabcd	0.040
<i>P. deltooides</i> Marsh × <i>P. simonii</i> Carr.	1.37 ± 0.37	Aab	0.47 ± 0.11	ABCabcd	0.055
<i>P. simonii</i> Carr. × <i>P. nigra</i> L.	1.32 ± 0.45	Aab	0.70 ± 0.16	ABab	0.154
<i>P. deltooides</i> Marsh × <i>P. cathayana</i> Rehd.	1.29 ± 0.35	Aab	0.80 ± 0.08	Aa	0.142
<i>P. deltooides</i> Marsh × <i>P. suaveolens</i> Fisch.	1.25 ± 0.23	Aab	0.63 ± 0.06	ABCabc	0.047
<i>P. ×euramericana</i> 'DN113'	1.15 ± 0.25	ABab	0.60 ± 0.12	ABCabcd	0.077
<i>P. alba</i> L. × <i>P. berlinensis</i> Dipp.	0.83 ± 0.30	ABbc	0.23 ± 0.09	BCcd	0.082
<i>P. pseudo-cathayana</i> × <i>P. deltooides</i> Barry 'Shan Hai Guan'	0.82 ± 0.20	ABbc	0.28 ± 0.06	BCbcd	0.047
<i>P. davidiana</i> Dode × <i>P. bolleana</i> Lauche.	0.80 ± 0.13	ABbc	0.29 ± 0.08	BCbcd	0.010
Dichloromethane (control)	0.17 ± 0.06	Bc	0.19 ± 0.07	Cd	0.689

Notes: Data in the table are mean ± SD, Tukey's multiple test and two-sample t tests

TABLE-2  
BEHAVIOURAL RESPONSE OF FEMALE AND MALE OF *S. populnea* ADULTS TO ESSENTIAL OIL OF 11 *Populus* VARIETIES

Essential oil type	Female				Male			
	Luring rate (%)	Repellent rate (%)	Response rate (%)	Paired t-test	Luring rate (%)	Repellent rate (%)	Response rate (%)	Paired t-test
<i>P. pseudo-simonii</i> Kitag. × <i>P. nigra</i> L.	85.0	10.0	95.0	0.006**	80.0	11.7	91.7	0.002**
<i>P. nigra</i> L.	68.3	21.7	90.0	0.002**	56.7	21.7	78.4	0.002**
<i>P. stalinetz</i> Jabl.	56.7	30.0	86.7	0.000**	48.3	36.7	85.0	0.020*
<i>P. deltooides</i> Marsh × <i>P. simonii</i> Carr.	68.3	26.7	95.0	0.001**	46.7	36.7	83.4	0.020*
<i>P. simonii</i> Carr. × <i>P. nigra</i> L.	78.3	11.7	90.0	0.008**	63.3	11.7	75.0	0.001**
<i>P. deltooides</i> Marsh × <i>P. cathayana</i> Rehd.	93.3	5.0	98.3	0.007**	63.3	13.3	76.6	0.003**
<i>P. deltooides</i> Marsh × <i>P. suaveolens</i> Fisch.	55.0	31.7	86.7	0.000**	46.7	35.0	81.7	0.050
<i>P. ×euramericana</i> 'DN113'	88.3	6.6	94.9	0.034*	56.7	31.7	88.4	0.073
<i>P. alba</i> L. × <i>P. berlinensis</i> Dipp.	46.7	43.3	90.0	0.048*	50.0	38.3	88.3	0.016*
<i>P. pseudo-cathayana</i> × <i>P. deltooides</i> Barry 'Shan Hai Guan'	56.7	30.0	86.7	0.000**	48.3	36.7	85.0	0.020*
<i>P. davidiana</i> Dode × <i>P. bolleana</i> Lauche.	43.3	41.7	85.0	0.064	31.7	33.3	65.0	0.120

Notes: "\*\*"P>0.05, "\*\*\*"P<0.01

*P. simonii* Carr. × *P. nigra* L. and *P. deltooides* Marsh × *P. cathayana* Rehd. volatile oils were extremely significantly different from the EAG response to dichloromethane ( $P < 0.01$ ); the EAG response to *P. pseudo-simonii* Kitag. × *P. nigra* L., *P. nigra* L., *P. stalinetz* Jabl., *P. deltooides* Marsh × *P. simonii* Carr. and *P. deltooides* Marsh × *P. suaveolens* Fisch. volatile oils were significantly different compared with the EAG response to control (dichloromethane) ( $P < 0.05$ ). The two-sample T-test showed that male and female *Saperda populnea* L. adults had extremely significantly different EAG responses to *P. nigra* L. ( $P < 0.01$ ) and significantly different EAG responses to *P. pseudo-simonii* Kitag. × *P. nigra* L., *P. stalinetz* Jabl., *P. deltooides* Marsh × *P. suaveolens* Fisch., *P. pseudo-cathayana* × *P. deltooides* Barry 'Shan Hai Guan' and *P. davidiana* Dode × *P. bolleana* Lauche. volatile oils ( $P < 0.05$ ).

"Y" olfactometer bioassay results and selective behavioural response of *Saperda populnea* L. to volatile oils. The results showed that paired T-test revealed significant differences in lure response and repellent response for females to *P. ×euramericana* 'DN113' and *P. alba* L. × *P. berlinensis* Dipp. volatile oils ( $P < 0.05$ ), extremely significant differences to *P. pseudo-simonii* Kitag. × *P. nigra* L., *P. nigra* L., *P. stalinetz* Jabl., *P. deltooides* Marsh × *P. simonii* Carr., *P. simonii* Carr. × *P. nigra* L., *P. deltooides* Marsh × *P. cathayana* Rehd., *P. deltooides* Marsh × *P. suaveolens* Fisch. and *P. pseudo-*

*cathayana* × *P. deltooides* Barry 'Shan Hai Guan' volatile oils ( $P < 0.01$ ) and no significant difference to *P. davidiana* Dode × *P. bolleana* Lauche. volatile oil ( $P > 0.05$ ). In males, paired T-test revealed significant differences in lure response and repellent response to *P. stalinetz* Jabl., *P. deltooides* Marsh × *P. simonii* Carr., *P. pseudo-cathayana* × *P. deltooides* Barry 'Shan Hai Guan' and *P. alba* L. × *P. berlinensis* Dipp. volatile oils ( $P < 0.05$ ), extremely significant differences to *P. pseudo-simonii* Kitag. × *P. nigra* L., *P. nigra* L., *P. simonii* Carr. × *P. nigra* L. and *P. deltooides* Marsh × *P. cathayana* Rehd. volatile oils ( $P < 0.01$ ) and no significant difference to *P. ×euramericana* 'DN113', *P. deltooides* Marsh × *P. suaveolens* Fisch. or *P. davidiana* Dode × *P. bolleana* Lauche. volatile oils ( $P > 0.05$ ) (Table-2).

*P. deltooides* Marsh × *P. cathayana* Rehd. displayed highest lure rate, 93.3 %, to female *Saperda populnea* L. and highest response rate. *P. pseudo-simonii* Kitag. × *P. nigra* L. had highest lure rate, 80.0 %, to male *Saperda populnea* L. and highest response rate. Overall, the response rate of female *Saperda populnea* L. was higher than male and the choice number to volatile oils in females was also higher than in males (Figs. 1 and 2).

**Composition and content of major volatiles in 11 kinds of poplar:** In volatile oils from branches in 11 kinds of poplar at dormant stage, identified 30 compounds accounted for

TABLE-3  
CHEMICAL COMPOSITION OF VOLATILES OBTAINED BY DISTILLATION METHOD  
FROM BRANCHES OF 11 POPLAR VARIETIES AT DORMANT PERIOD

Compound name	Relative content (%)										
	A	B	C	D	E	F	G	H	I	J	K
2-Methyl-3-buten-2-ol	6.27	2.25	22.43	3.10	2.43	2.44	3.15	5.18		0.99	
3-Methyl-2-buten-1-ol	9.28	5.56		1.71	4.90	2.28	6.18	7.03			
2-Methyl-2-buten-1-ol			16.15	2.01		2.14					
<i>p</i> -Xylene		0.44		0.66	2.28		6.45		0.69		2.49
Formaldehyde		0.32		0.74		0.37					
2-Hydroxy-cyclohexanone	3.04	0.66	0.39	2.25	3.79	12.97		0.31	1.72	15.34	
phenol	4.99	4.55	5.15	4.36	1.07	0.85	1.54	2.03	6.71	3.89	2.91
1,2-Cyclohexanedione	22.42	23.18	5.54	21.87	15.54	39.62	10.70	9.63	51.55	36.75	21.74
2-Methyl-4-methylene-hexane	3.22			1.41							2.88
2-Cyclohexen-1-one	12.11	13.95	3.29	9.89	11.81	6.83	3.40	3.32	18.32	7.89	12.72
2-Hydroxy-benzaldehyde		0.45	5.50	16.32	2.90	0.70	5.72	1.19	4.70		1.43
3,7-Dimethyl-1,6-sim-3-ol		2.60	1.07	0.63		0.29		5.27		3.08	2.32
Phenylethanol	3.04	4.23	4.02	2.66	2.45	0.24	1.54	1.12	1.33	1.03	6.58
Borneol				5.23		0.37					
2,3-Dihydrobenzofuran						0.20			1.03		
Eugenol	2.15		3.70	0.74		0.38			0.70		
Ylangene		0.47				0.79	0.53	0.77			
Caryophyllene		0.35	0.84					0.74			
$\delta$ -Selinene		2.56	6.82	3.19				10.69	1.09		
Toluene		0.49		0.51	5.50	0.28			0.47	0.32	1.45
Furfural			0.73	2.05		0.31		0.30		0.29	
Benzyl alcohol	1.62	4.71	1.95	3.74		1.32	10.06		2.66	2.04	
$\alpha$ -Bisabolol	1.70			0.54			10.94	0.61			6.64
Terpane						2.27		13.14		1.79	
(2 <i>R</i> - <i>cis</i> )-1,2,3,4,4 $\alpha$ , 5,6,7-octahydro- $\alpha$ , $\alpha$ , 4 $\alpha$ , 8-tetramethyl-2--naphthalene methanol	6.70			3.49	8.19			7.98			
2-Naphthalenemethanol, decahydro- $\alpha$ , $\alpha$ , 4 $\alpha$ -trimethyl-8-methylene-, [2 <i>R</i> -(2 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\beta$ )]-	7.67	8.32	5.70	2.83	9.80	10.12	10.94	19.94	2.05	11.63	16.73
2-Naphthalenemethanol, octahydro- $\alpha$ , $\alpha$ , 4 $\alpha$ -tetramethyl-, [2 <i>R</i> -(2 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\beta$ )]-			6.83		11.37					0.32	
$\alpha$ -Cedrene	4.13	2.45	1.59	3.81			22.80	1.28			15.50
Bicyclo[3.1.1]hept-2-ene, 2,6-dimethyl-6-(4-methyl-3-pentenyl)-	0.94	6.75		0.85	8.32	6.87	5.94			3.04	5.24
Naphthalene, 1,2,3,4, 4a, 5,6, 8a-octahydro-7-methyl-4-methylene-1-(1-methylethyl)-, (1 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\beta$ )-			0.88								
Total	89.28	84.29	92.58	94.59	90.35	91.64	96.74	90.53	93.02	88.40	98.63

90.35, 84.29, 92.58, 91.64, 94.59, 90.53, 89.28, 93.02, 93.02, 98.63 and 88.40 % of volatile oil total content, respectively. Among them, the common compounds included 1,2-cyclohexanedione, 2-cyclohexene-1-one, phenol, benzene, ethanol and [2*R*-(2 $\alpha$ , 4 $\alpha$ ,  $\alpha$ , 8 $\alpha$ ,  $\beta$ )]-decahydro- $\alpha$ ,  $\alpha$ , 4 $\alpha$ ,-three

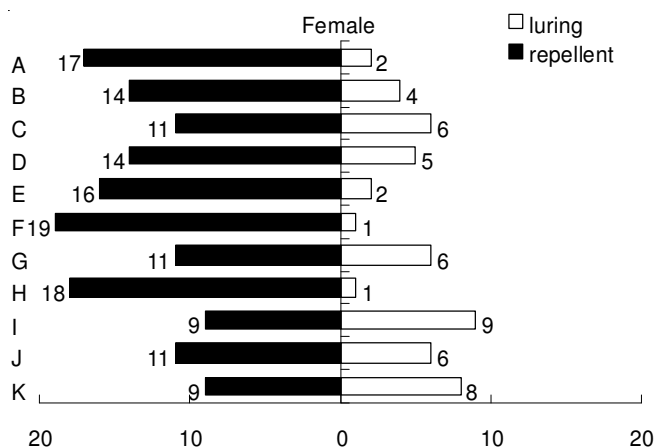


Fig. 1. Selective response of female of *S. populnea* adults to essential oil of 11 *Populus* varieties

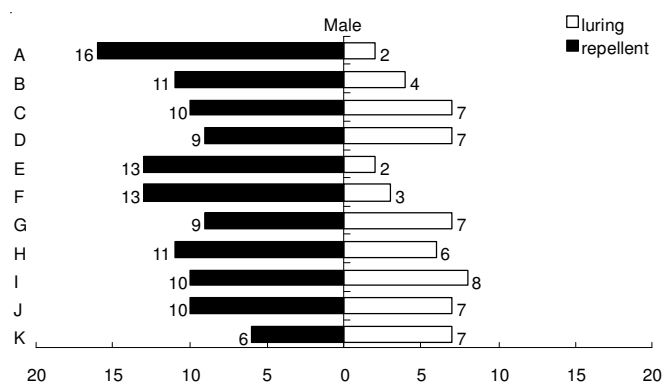


Fig. 2. Selective response of male of *S. populnea* adults to essential oil of 11 *Populus* varieties

methyl-8-methene-2-naphthalene methanol. 1,2-cyclohexanedione had the highest content in branches of *P. simonii* Carr.  $\times$  *P. nigra* L., *P. nigra* L., *P. deltoides* Marsh  $\times$  *P. cathayana* Rehd., *P. deltoides* Marsh  $\times$  *P. simonii* Carr., *P. pseudo-simonii* Kitag.  $\times$  *P. nigra* L., *P. alba* L.  $\times$  *P. berolinensis* Dipp., *P. davidiana* Dode  $\times$  *P. bolleana* Lauche and *P. pseudo-cathayana*  $\times$  *P. deltoides* Barry 'Shan Hai Guan'; 2-methyl-3-



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2-Methyl-2-buten-1-ol			16.15	2.01		2.14					
<i>p</i> -Xylene		0.44		0.66	2.28		6.45		0.69		2.49
Formaldehyde		0.32		0.74		0.37					
2-Hydroxy-cyclohexanone	3.04	0.66	0.39	2.25	3.79	12.97		0.31	1.72	15.34	
phenol	4.99	4.55	5.15	4.36	1.07	0.85	1.54	2.03	6.71	3.89	2.91
1,2-Cyclohexanedione	22.42	23.18	5.54	21.87	15.54	39.62	10.70	9.63	51.55	36.75	21.74
2-Methyl-4-methylene-hexane	3.22			1.41							2.88
2-Cyclohexen-1-one	12.11	13.95	3.29	9.89	11.81	6.83	3.40	3.32	18.32	7.89	12.72
2-Hydroxy-benzaldehyde		0.45	5.50	16.32	2.90	0.70	5.72	1.19	4.70		1.43
3,7-Dimethyl-1,6-sim-3-ol		2.60	1.07	0.63		0.29		5.27		3.08	2.32
Phenylethanol	3.04	4.23	4.02	2.66	2.45	0.24	1.54	1.12	1.33	1.03	6.58
Borneol				5.23		0.37					
2,3-Dihydrobenzofuran						0.20			1.03		
Eugenol	2.15		3.70	0.74		0.38			0.70		
Ylangene		0.47				0.79	0.53	0.77			
Caryophyllene		0.35	0.84					0.74			
$\delta$ -Selinene		2.56	6.82	3.19				10.69	1.09		
Toluene		0.49		0.51	5.50	0.28			0.47	0.32	1.45
Furfural			0.73	2.05		0.31		0.30		0.29	
Benzyl alcohol	1.62	4.71	1.95	3.74		1.32	10.06		2.66	2.04	
$\alpha$ -Bisabolol	1.70			0.54			10.94	0.61			6.64
Terpane						2.27		13.14		1.79	
(2 <i>R</i> - <i>cis</i> )-1,2,3,4,4 $\alpha$ , 5,6,7-octahydro- $\alpha$ , $\alpha$ , 4 $\alpha$ , 8 – tetramethyl-2-- naphthalene methanol	6.70			3.49	8.19			7.98			
2-Naphthalenemethanol, decahydro- $\alpha$ , $\alpha$ , 4 $\alpha$ -trimethyl-8-methylene-, [2 <i>R</i> -(2 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\beta$ )]-	7.67	8.32	5.70	2.83	9.80	10.12	10.94	19.94	2.05	11.63	16.73
2-Naphthalenemethanol, octahydro- $\alpha$ , $\alpha$ , 4 $\alpha$ -tetramethyl-, [2 <i>R</i> -(2 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\beta$ )]-			6.83		11.37					0.32	
$\alpha$ -Cedrene	4.13	2.45	1.59	3.81			22.80	1.28			15.50
Bicyclo[3.1.1]hept-2-ene, 2,6-dimethyl-6-(4-methyl-3-pentenyl)-	0.94	6.75		0.85	8.32	6.87	5.94			3.04	5.24
Naphthalene, 1,2,3,4, 4a, 5,6, 8a-octahydro-7-methyl-4-methylene-1-(1-methylethyl)-, (1 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\beta$ )-			0.88								
Total	89.28	84.29	92.58	94.59	90.35	91.64	96.74	90.53	93.02	88.40	98.63

buten-2-ol had the highest content in branches of *P. stalinetz* Jabl.; [2*R*-(2 $\alpha$ , 4 $\alpha$ ,  $\alpha$ , 8 $\alpha$ ,  $\beta$ )]-decahydro- $\alpha$ ,  $\alpha$ , 4 $\alpha$ ,-three methyl-8-methene-2-naphthalene methanol had the highest content in branches of *P.  $\times$  euramericana* 'DN113';  $\alpha$ -Cedrene had the highest content in branches of *P. deltooides* Marsh  $\times$  *P. suaveolens* Fisch. (Table-3).

In volatile oils from branches in 11 kinds of poplar at growth stage, identified 35 compounds accounted for 97.75, 99.96, 85.43, 85.04, 99.57, 97.82, 94.06, 92.63, 99.99, 99.13 and 99.92 %, of volatile oil total content, respectively. Among them, the common compounds included *p*-xylene, 1,2-cyclohexanedione, 2-cyclohexene-1-one and 7-ethoxy coumarin. 1,2-Cyclohexanedione had the highest content in branches of *P. simonii* Carr.  $\times$  *P. nigra* L., *P. deltooides* Marsh  $\times$  *P. cathayana* Rehd., *P. alba* L.  $\times$  *P. berlinensis* Dipp., *P. davidiana* Dode  $\times$  *P. bolleana* Lauche and *P. pseudo-cathayana*  $\times$  *P. deltooides* Barry 'Shan Hai Guan'; *p*-xylene had the highest content in branches of *P. nigra* L., *P. stalinetz* Jabl., *P.  $\times$  euramericana* 'DN113' and *P. deltooides* Marsh  $\times$  *P. suaveolens* Fisch; 2-hydroxybenzaldehyde had the highest content in branches of *P. deltooides* Marsh  $\times$  *P. simonii* Carr.;

eugenol ad the highest content in branches of *P. pseudo-simonii* Kitag.  $\times$  *P. nigra* L. (Table-4).

Comparison of growth stage and dormant stage identified 18 kinds of same compounds, among which 1,2-cyclohexanedione and 2-cyclohexene-1-one were present in branches from all the 11 types of poplars at both growth stage and dormant stage. The six kinds of aromatic compounds in volatile oil from branches at dormant stage were: *p*-xylene, phenol, 2-hydroxy-benzene, phenethyl alcohol, toluene and benzil alcohol; there were six kinds of aromatic compounds in volatile oil from branches at growth stage. including *p*-xylene, phenol, toluene, 2-hydroxy-benzene, ethyl benzene and 1,2,3-trimethylbenzene. In the 11 kinds of poplars, only *P. alba* L.  $\times$  *P. berlinensis* Dipp. and *P. davidiana* Dode  $\times$  *P. bolleana* Lauche. had lower number of aromatic compounds at growth stage than at dormant stage.

EAG results showed that female and male *Saperda populnea* L. adults had strong response activity to volatile oils from branches of *P. pseudo-simonii* Kitag.  $\times$  *P. nigra* L. (1.70  $\pm$  0.25, 0.55  $\pm$  0.23), *P. simonii* Carr.  $\times$  *P. nigra* L. (1.32  $\pm$  0.45, 0.70  $\pm$  0.16), *P. deltooides* Marsh  $\times$  *P. cathayana* Rehd.

TABLE-4  
CHEMICAL COMPOSITION OF VOLATILES OBTAINED BY DISTILLATION METHOD  
FROM BRANCHES OF 11 *POPLAR* VARIETIES AT GROWING PERIOD

Compound name	Relative content (%)										
	A	B	C	D	E	F	G	H	I	J	K
Toluene						0.33	0.43		0.18		
2-Methyl-2-buten-1-ol			0.88								
Acetaldehyde				0.86		0.47	1.09				1.87
Phenylethane			1.94	0.79	1.47	1.16	2.48	2.32	0.63		2.57
Xylene	4.85	43.31	24.09	9.13	25.64	14.06	29.65	29.22	9.26	0.72	36.30
1,2,3-trimethyl- benzene		14.91	0.54	2.97	0.46	0.28	1.5	0.52	0.26		0.58
Phenol									1.01		
2-Hydroxy-cyclohexanone						0.21					
1,2-Cyclohexanedione	11.06	13.81	8.78	1.12	36.02	17.64	20.37	19.40	37.66	26.46	20.61
2-Cyclohexen-1-one	7.02	4.78	4.70	0.52	17.29	11.10	12.72	2.70	31.01	14.53	20.05
Terpane				2.26				10.00			
2-Hydroxy-benzaldehyde	11.28		4.74	68.42	3.82	3.27	2.36				
Borneol		1.25		6.96		1.01		1.43			
7-Ethoxycoumarin	3.46	14.91	13.15	4.47	11.71	5.78	13.74	13.43	4.35	0.85	15.41
2-Sec-Buthlphenol			2.14								
Tridecane									0.58		
Copaene	7.10	1.49				0.78					
Bicyclo[3.1.1]hept-2-ene, 2,6-dimethyl-6-(4-methyl-3-pentenyl)-						0.65					
Eugenol	17.51		15.53								
Naphthalene-(1S)-1,2,3,5,6,8 $\alpha$ -Hexahydro-4,7-1-(1-methyl-ethyl)											
Naphthalene-1,2,3,4-tetrahydro-1,6-dimethyl-4-(1-methyl-ethyl)											1.52
Caryophyllene						0.64	3.21		0.75	0.94	
[9.1.0]Dodecane-3,7-diene							1.68				
Decahydro-1,1,7-trimethyl-4-cyproterone alkenyl-1H-[e]							1.12				1.80
Ylangene				0.21						0.46	1.48
Naphthalene-[1 $\alpha$ R-(1 $\alpha$ , $\alpha$ , 3 $\alpha$ , $\alpha$ , 7 $\beta$ , $\alpha$ )]-1 $\alpha$ , 2, 3, 3 $\alpha$ , 4, 5, 6, 7 $\beta$ -octahydro-1, 1, 3 $\alpha$ , 7-tetramethyl-hydroge cyclopropyl	1.45								0.25	0.99	
Naphthalene-(1 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\alpha$ )-1, 2, 4 $\alpha$ , 5, 6, 8 $\alpha$ -hexahydro-4,7-dimethyl-1-(1-methyl-ethyl)			1.11			2.02	2.51	1.72			1.97
Naphthalene-(1 $\alpha$ , 4 $\alpha$ , $\beta$ , 8 $\alpha$ , $\alpha$ )-1, 2, 3, 4, 4 $\alpha$ , 5, 6, 8 $\alpha$ -octahydro-7-methyl-4methylene-1-(1-methyl-ethyl)		2.27	7.83	1.86		1.98	1.34	9.06			1.41
[2R-(2 $\alpha$ , 4 $\alpha$ , $\alpha$ , 8 $\alpha$ , $\beta$ )]-decahydro- $\alpha$ , 4 $\alpha$ , -trimethyl-8-methylene-2-naphthalene methanol					1.34	3.27	2.43	2.38	0.60		
1, 2, 3, 3 $\alpha$ , 4, 5, 6, 7, 8-octahydro- $\alpha$ , $\alpha$ , 3, 8-tetramethyl-1,5-chamomile central methanol	17.10	3.23				3.87	3.62		1.86	20.00	
$\alpha$ -Humulene											6.09
$\alpha$ -Bisabolol											1.17
4, 8 $\alpha$ -Dimethyl-6-different allyl-1, 2, 3, 5, 6, 7, 8, 8 $\alpha$ -octahydro-naphthalene-2-ol						15.40		5.64	3.77	19.99	2.60
Linoleic acid	6.78										
Linolenic acid	6.45										
Total	94.06	99.96	85.43	99.57	97.75	85.04	99.13	97.82	92.63	99.92	99.99

(1.29  $\pm$  0.35, 0.80  $\pm$  0.08) and *P. nigra* L. (1.58  $\pm$  0.20, 0.63  $\pm$  0.17), but weak response to volatile oils from branches of *P. davidiana* Dode  $\times$  *P. bolleana* Lauche (0.80  $\pm$  0.13, 0.29  $\pm$  0.08), *P. alba* L.  $\times$  *P. berolinensis* Dipp. (0.83  $\pm$  0.30, 0.23  $\pm$  0.09) and *P. pseudo-cathayana*  $\times$  *P. deltooides* Barry 'Shan Hai Guan' (0.82  $\pm$  0.20, 0.28  $\pm$  0.06). "Y" type olfactometer measurement showed that volatile oils from branches of *Populus nigra*, including *P. simonii* Carr.  $\times$  *P. nigra* L. (78.3 %, 63.3 %), *P. deltooides* Marsh  $\times$  *P. cathayana* Rehd. (93.3 %, 63.3 %), *P.  $\times$  euramericana* 'DN113' (88.3 %, 56.7 %) and *P. pseudo-simonii* Kitag.  $\times$  *P. nigra* L. (85.0 %, 80.0 %), had strong lure effects to female and male *Saperda populnea* L. adults, while volatile oils from branches of *P. davidiana*

Dode  $\times$  *P. bolleana* Lauche (43.3 %, 31.7 %) showed strong resistance. This was consistent with the field survey results that *Populus nigra* was seriously damaged by *Saperda populnea* L.<sup>16</sup>. In addition, we also found that the EAG responses of female *Saperda populnea* L. were higher than males, so did the selection rate on "Y" type olfactometer and there were large difference between females and males. Thus, we believe that female *Saperda populnea* L. had more sensitive olfactory than males. This was also proved by the fact that the "A-3 Monochamus alternates attractant", a new product developed by Guangdong Forestry Research Institute, *etc.*<sup>17</sup>, lured more female *Monochamus alternates* than males.

The chemical composition and relative content of volatile oils at different stages showed complex composition of volatile oils. The main ingredients were alcohols, ketones, alkanes, terpenes and heterocyclic compounds. 1,2-Cyclohexanedione, 2-cyclohexene-1-one and aromatic compounds were major compounds of volatile oils from poplar branches, but the relative contents differed in different species at different stages. Less xylene existed at branches from different poplars at dormant stage, but xylene was widely and extensively present at growth stage. The fatty alcohols (2-methyl-3-buten-2 alcohol and 2-methyl -2-butene-1-ol) in volatile oil from dominant branch was proved to be derivatives of semi-terpenoids sorbitol and isopentenyl diphosphate. It exists in poplar shoots<sup>18</sup>, but the content is extremely low in volatile oil from branches at growth state; it is a synthesized compound during plant growth and development. The chemical composition and relative content of plant volatile oil not only depend on soil composition, climatic condition, growing area, season, air and soil humidity and age, but also depend on the extraction method, extraction conditions and extraction reagents. Furthermore, different physiological states, such as human physical damage, pests and diseases, also induce plants to initiatively synthesize and release new volatile compounds, mainly terpenoids. For example, *P. pseudo-simonii* Kitag. × *P. nigra* L. damaged by *Quadraspidiotus gigas* Thiem et Gerneck displayed strong luring effect to *Pteroptrix longgiclava* Girault and *Encarsia gigas* Tshumakora<sup>19</sup>.

According to the GC-MS results and composition analysis of some *Cerambycidae* attractants that have been applied, the preliminary conclusion is that the effective *Saperda populnea* L. attractant is a mixture of components. As identified by GC-MS in this study, the common compounds in volatile oils from branches of 11 poplars were 1,2-cyclohexanedione, 2-cyclohexene-1-one and aromatic compounds. Combined with previous studies, we believe that these two compounds are components of specific smell in poplar branch volatile, while phenethyl alcohol, phenol and other aromatic compounds are host odor with lure effects. Lampman *et al.*<sup>20</sup> reported that benzaldehyde, eugenol and other aromatic compounds had strong lure activity to *Diabrotica virgifera* Leconte adults; Heath *et al.*<sup>21</sup> showed that jasmine released phenylacetaldehyde had obvious lure effects to *Trichoplusiani*. Host plant odors

play key roles in inducing herbivorous insects to feed, spawn, *etc.* Therefore, attractants with effective lure activity to *Saperda populnea* L. females can be screened out using phenylacetaldehyde, phenol and other aromatic compound monomers, supplementing with different concentrations of 1,2-cyclohexanedione and 2-cyclohexene-1-one, with *Saperda populnea* L. female as targets and through a large number of biometric screening. Its application in forest can change the ratio of male and female adults by trapping *Saperda populnea* L. females to effectively control the female population, thereby reducing *Saperda populnea* L. damage.

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