

# Species and quantity of airborne pollens in Shanghai as monitored by gravitational and volumetric methods

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## Abstract

**Background:** The prevalence of allergic diseases has markedly increased in the last decades. It is therefore important to assess the distribution of airborne pollen, the most important aeroallergen, for allergic disease prevention and control.

**Objective:** To identify the species and quantity of airborne pollens, and observe their distribution characteristics in Shanghai, using gravitational (Durham Sampler) and volumetric (Rotorod Sampler 40) methods simultaneously. In addition, the correlation between both methods was analyzed to provide effective preventive measures for pollen-sensitized individuals.

**Method:** Pollen counts were monitored in the same area from November 1, 2009 to October 31, 2010 by samplers set at the same height and site. Pollen concentrations as well as any association between the two methods were determined.

**Results:** Two pollen concentration peaks in Shanghai were observed from March to May (spring) and September to October (autumn). In spring, tree pollen was the main species, with a predominance of *Broussonetia*. In autumn, grass pollen predominated, with mostly *Humulus*. Thirty-two species were identified by both gravitational and volumetric methods. Five and seven additional species were identified exclusively by the gravitational and volumetric methods, respectively. Pollen counts obtained from both devices were significantly correlated ( $P < 0.05$ ).

**Conclusions:** Two methods were used simultaneously for the first time to monitor pollen counts in central urban Shanghai, showing two annual peaks. *Broussonetia* and *Humulus* were the predominant spring and autumn pollens, respectively. Pollen counts obtained by both methods were clearly correlated. Regional airborne pollen monitoring offers preventive measures for sensitized individuals and provides useful clinical information.

**Keywords:** pollen count; Durham sampler; Rotorod sampler 40; gravitational method; volumetric method

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## Introduction

The prevalence of allergic diseases has markedly increased.<sup>1-4</sup> Multiple airborne pollen species are important bio-pollutants responsible for human respiratory allergies.

Airborne pollen is the most important aeroallergen in outdoor air, and can release numerous soluble proteins<sup>5</sup> after being drawn into the nasal cavity and bronchial tree with each breath; this can lead to seasonal allergic rhinitis and asthma, with the potential to induce allergic sensitization.<sup>6,7</sup> The current prevalence of pollen hypersensitivity is on the rise due to

significantly increased airborne pollen counts of disparate species, which is due in part to rapid sustainable economic development in China, accelerated urban greening, changes in the types of crops cultivated, windbreak and sand fixation, and the invasion of harmful exotic plants. Differences exist among pollen species regarding their relative counts and seasonal distributions in China's vast territory. There have been few reports assessing airborne pollens in Shanghai;<sup>8,9</sup> thus, it is important to evaluate the distribution of airborne pollens in

ambient air for the prevention and control of allergic diseases in the Chinese population.

Several Chinese scholars have used the gravitational method to monitor airborne pollens since the early 1950s, and assessed pollen distributions throughout the country in the 1980s to determine the contribution of pollen species for the first time, accumulating valuable information.<sup>10</sup> The gravitational method is inexpensive and easy to promote; in addition, it provides important guidance with respect to the clinical management of allergic airway diseases. The Durham sampler is commonly used in China at this time. However, only pollen counts per unit area ( $\text{g}/1000 \text{ mm}^2$ ) can be evaluated by this method, not the actual pollen concentrations.<sup>11</sup> This means that pollen counts are given per cubic meter,  $\text{g}/\text{m}^3$ , which is directly related to the severity of hay fever.<sup>12-14</sup>

The Rotorod sampler 40 is recognized internationally, with certain advantages compared with the gravitational method. It is based on the principles of aerodynamics and determines pollen counts per cubic meter in air, thereby providing actual pollen concentrations.<sup>15,16</sup>

The current study monitored ambient airborne pollen counts from November 2009 to October 2010, using both the gravitational method and the Rotorod sampler 40 simultaneously; in addition, the correlation between the two methods was assessed.

## Methods

### Key instruments

The Durham sampler was manufactured by the Department of Allergy, Xiehe Hospital (Beijing); the Rotorod sampler 40 was obtained from Sampling Technologies, Inc. (USA). The microscope and ocular micrometer of the SE type were produced by Nikon (Japan).

### Main reagents and consumables

White petroleum jelly (Sinopharm Chemical Reagent Co., Ltd), glycerin (Shenyang TianGang Chemical Reagent), agar powder (Beijing Yili Fine Chemicals Co., Ltd), anhydrous ethanol (Changzhou WuWei Reagent Co., Ltd), fuchsine (Sinopharm Shanghai Chemical Reagent) and slides (Shanghai Biaoyi Instrument Co., Ltd) were used in this study.

Calberla's dye, silicone oil, and 1.52x1.52x32 mm polystyrene pollen sampling rods were purchased from Sampling Technologies, Inc. (USA).

### Sampler positioning

Two samplers were placed on the roof of the ward building, located about 15 meters high, at Shanghai Renji Hospital, which is central to the Huangpu district in Shanghai, China; this location can be considered the center of Shanghai. Samplers were fixed on a bracket about 1.5 meters above ground, with no tall buildings nearby, and positioned away from air conditioning units and vegetation. The Rotorod sampler was installed in accordance with the manufacturer's instructions, placed about 1.5 meters away from the Durham sampler, with a long-term power supply. The collection time setting was a 5% duty cycle (i.e., in a period of 600 sec, the sampling rod rotated for 30 sec and remained still for 9 min and 30 sec).

### Pollen collection

Slides and sampler rods were placed at the same time (8:00 A.M.), recycled, stained and counted before the next 8:00 A.M. sampling cycle, when new slides and sample rods were placed. Microscopes and an SE type ocular micrometer were used to identify pollen species by qualified botanists at East China Normal University. Pollens were identified at the genus level, or at the family level if only minimal differences were found among the identified genera.

### Statistical analysis

Excel 2003 was applied to establish archived databases, and the SPSS version 13.0 statistical software was used for data processing. The correlation of pollen count monitoring between the two methods was assessed by Pearson correlation coefficient ( $r$ ).  $P < 0.05$  was considered statistically significant.

## Results

### Data obtained by the gravitational method using the Durham sampler

#### Pollen counts

A total of 37 pollen species amounting to  $12046 \text{ g}/\text{m}^2$  were identified from November 1, 2009 to October 31, 2010; among them, the nine main species were *Broussonetia*, *Humulus*, *Gramineae*, *Pinaceae*, *Platanus*, *Taxodiaceae*, *Cupressaceae*, *Ginkgo biloba* and *Betulaceae* in order of significance (Table 1). As shown in Figure 1A, airborne pollen counts peaked from March to May, with *Broussonetia*, *Pinaceae* and *Platanus* as the most abundant groups. There was a small peak from

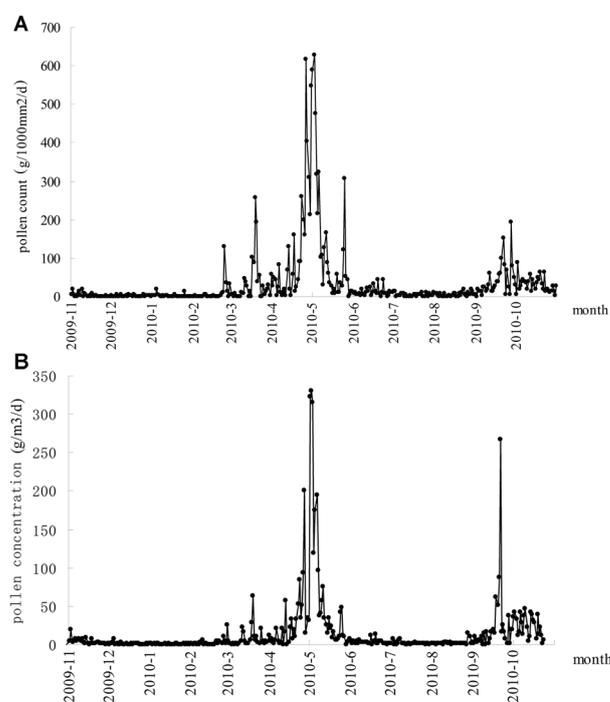


Figure 1. A, Distribution of airborne pollen counts in the central urban area of Shanghai from November 2009 to October 2010. B, Distribution of airborne pollen concentrations in central urban area of Shanghai from November 2009 to October 2010.

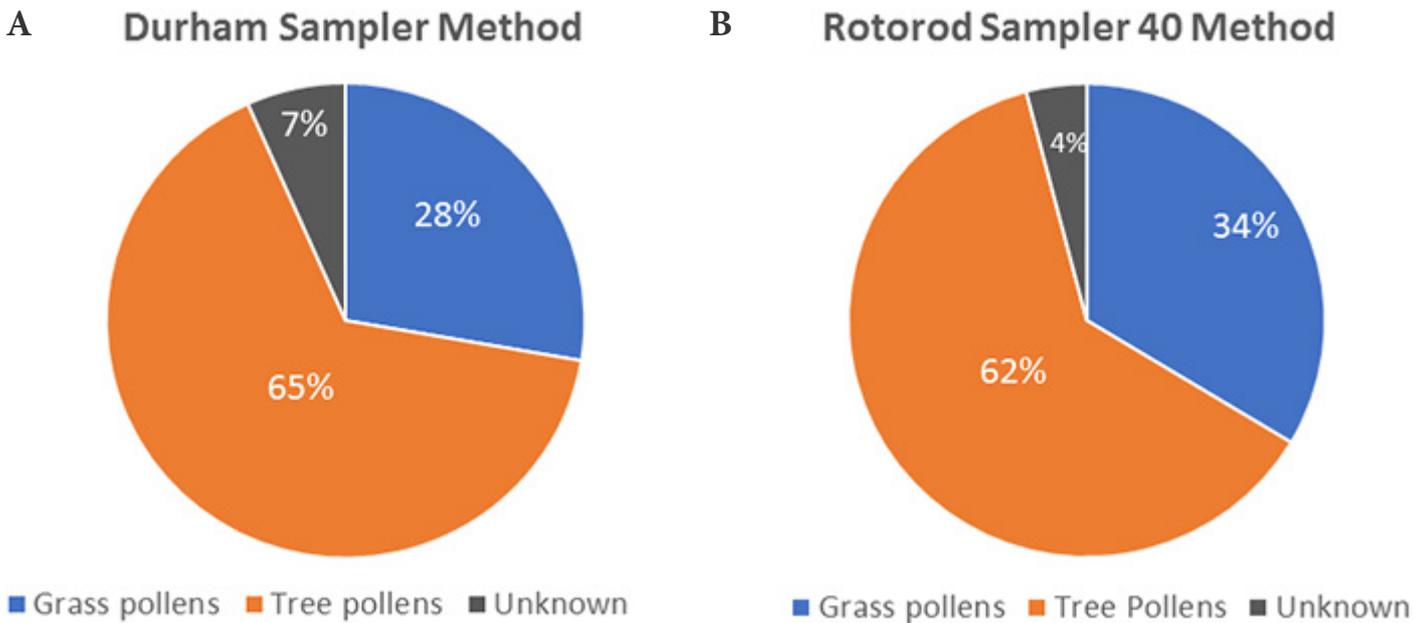


Figure 2. A, Composition of airborne pollen counts in the central urban area of Shanghai from November 2009 to October 2010, as assessed by gravitational method with Durham Sampler. B, Composition of airborne pollen concentrations in the central urban area of Shanghai from November 2009 to October 2010, as assessed by the volumetric method with Rotorod Sampler 40.

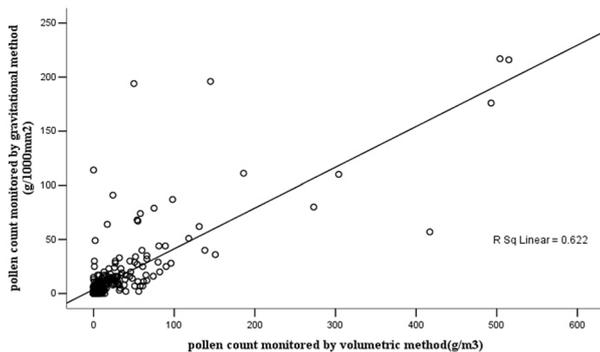


Figure 3. Correlation of pollen counts as monitored by the gravitational and volumetric methods.

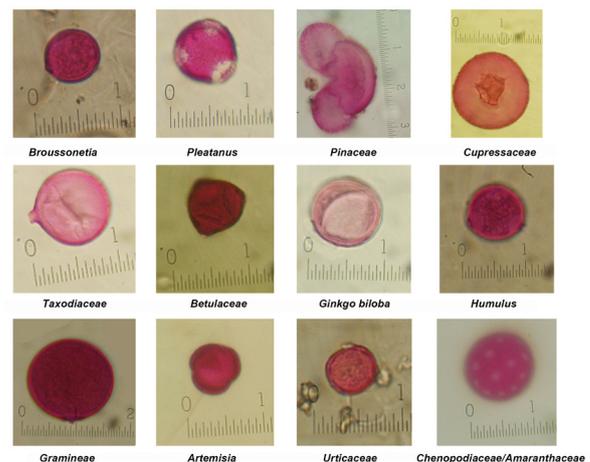


Figure 4. Images of several main pollen species.

September to October, with *Humulus* and *Gramineae* as the dominant groups. The highest daily pollen counts in spring and autumn were 628 g/1000 mm<sup>2</sup>/d and 193 g/1000 mm<sup>2</sup>/d, respectively. In addition, *Gramineae* was found throughout the year.

#### Pollen species composition

There were obvious differences in the pollen species distribution throughout the year. Tree pollen types were predominantly found in spring. A total of 26 pollen species, amounting to 8600 g/1000 mm<sup>2</sup>, were identified from March 1 to May 31, 2010, and constituted 71.36% of the overall pollen count. The most abundant species were *Broussonetia* (45.64%), *Pinaceae* (7.86%), *Platanus* (6.06%), *Gramineae* (4.60%) and *Cupressaceae* (4.55%). Grass pollens were predominant in autumn. A total of 15 pollen species, amounting to 2276 g/1000 mm<sup>2</sup>, were identified from September 1 to October 31, 2010, comprising 18.89% of the overall pollen count. The most abundant species were *Humulus* (59.01%), *Gramineae* (18.45%), *Artemisia* (5.32%), *Chenopodiaceae/Amaranthaceae* (4.17%),

and *Compositae* (3.43%). Among them, tree pollens and grass pollens comprised 65.55% and 27.74% of the overall pollen count, respectively (Figure 2A).

#### Data obtained by the volumetric method using the Rotorod sampler 40

##### Pollen concentration

A total of 39 pollen species, amounting to 4818 g/m<sup>3</sup>, were identified from November 1, 2009 to October 31, 2010, with more than ten pollen species in high concentrations. *Chenopodiaceae* and *Amaranthaceae* were combined in the analysis, due to their similar configurations (see Table 1). Airborne pollen concentrations peaked from March to May, with a small peak observed from September to October (Figure 1B). The highest daily concentrations in spring and autumn were 330 g/m<sup>3</sup>/d and 267 g/m<sup>3</sup>/d, respectively. *Gramineae* showed a biphasic peak in May and September to October.

**Table 1. Monthly distribution of airborne pollen counts in the central urban area of Shanghai by the gravitational method with Durham Sampler**

Pollen species	Pollen count (g/1000mm <sup>2</sup> )													Proportion of all (%)
	Nov in 2009	Dec in 2009	Jan in 2010	Feb in 2010	Mar in 2010	Apr in 2010	May in 2010	Jun in 2010	Jul in 2010	Aug in 2010	Sep in 2010	Oct in 2010	Sum	
1. <i>Broussonetia</i>	0	0	0	0	0	1230	2695	5	0	3	0	0	3933	32.64
2. <i>Humulus</i>	23	8	0	0	0	0	0	0	0	18	1018	325	1392	11.55
3. <i>Gramineae</i>	10	5	0	3	28	148	220	45	48	45	135	285	972	8.07
4. <i>Pinaceae</i>	0	0	0	5	88	253	335	18	5	0	0	3	707	5.87
5. <i>Platanus</i>	0	0	0	0	0	493	28	0	0	0	0	0	521	4.32
6. <i>Taxodiaceae</i>	0	0	3	145	200	108	0	5	8	0	0	0	469	3.89
7. <i>Cupressaceae</i>	0	0	0	70	378	8	5	0	0	0	0	0	461	3.83
8. <i>Ginkgo biloba</i>	0	0	0	0	5	278	0	0	0	0	0	0	283	2.35
9. <i>Betulaceae</i>	5	3	0	3	43	103	88	5	5	3	0	0	258	2.14
10. <i>Chenopodiaceae/ Amaranthaceae</i>	5	3	18	0	5	18	30	10	10	23	65	30	217	1.80
11. <i>Ulmaceae</i>	0	3	3	15	70	113	10	0	0	3	0	0	217	1.80
12. <i>Moraceae</i>	0	0	0	0	13	140	23	3	5	5	0	0	189	1.57
13. <i>Artemisia</i>	13	43	3	0	0	0	0	0	0	5	48	73	185	1.54
14. <i>Cruciferae</i>	0	0	10	0	0	158	15	0	0	0	0	0	183	1.52
15. <i>Ailanthus</i>	0	0	0	0	0	130	25	0	0	0	0	0	155	1.29
16. <i>Compositae</i>	15	0	0	3	0	0	45	3	0	10	10	68	154	1.28
17. <i>Pterocarya</i>	0	0	3	0	8	123	8	0	0	0	0	0	142	1.18
18. <i>Paulownia</i>	0	0	0	0	3	45	88	0	0	0	0	0	136	1.13
19. <i>Palmae</i>	0	0	0	0	0	0	0	105	13	0	0	0	118	0.98
20. <i>Myrtle</i>	0	0	0	0	0	0	58	0	3	33	18	0	112	0.93
21. <i>Salix</i>	0	0	0	0	105	5	0	0	0	0	0	0	110	0.91
22. <i>Legume</i>	0	0	0	0	0	3	3	50	0	0	0	0	56	0.46
23. <i>Rosaceae</i>	0	0	0	0	8	33	10	0	0	0	0	0	51	0.42
24. <i>Populus</i>	0	0	0	0	5	40	3	0	0	0	0	0	48	0.40
25. <i>Plantaginaceae</i>	5	0	0	0	0	0	0	0	0	0	0	38	43	0.36
26. <i>Ricinus</i>	5	0	0	0	0	3	0	0	0	0	0	20	28	0.23
27. <i>Cyperaceae</i>	0	0	3	0	0	3	15	0	0	0	0	3	24	0.20
28. <i>Dianthus</i>	0	0	0	0	23	0	0	0	0	0	0	0	23	0.19
29. <i>Ericaceae</i>	3	0	0	0	0	0	0	0	0	0	0	13	16	0.13
30. <i>Rubiaceae</i>	0	0	0	0	0	0	0	0	0	0	0	10	10	0.08
31. <i>Koelreuteria</i>	0	0	0	0	0	0	0	0	0	0	8	0	8	0.07
32. <i>Ligustrum</i>	0	0	0	0	0	0	0	5	0	0	0	0	5	0.04
33. <i>Campanulaceae</i>	0	0	0	0	0	0	0	0	0	0	5	0	5	0.04
34. <i>Ragweed</i>	0	0	0	0	0	0	0	0	0	5	0	0	5	0.04
35. <i>Euphorbiaceae</i>	0	0	0	3	0	0	0	0	0	0	0	0	3	0.02
36. <i>Buxus</i>	0	0	0	0	3	0	0	0	0	0	0	0	3	0.02
36. unknown	15	20	43	3	50	188	238	98	38	15	8	93	809	6.71

**Table 2. Monthly distribution of airborne pollen concentrations in the central urban area of Shanghai by the volumetric method with Rotorod Sampler 40**

Pollen species	Pollen concentration(g/m <sup>3</sup> )													Proportion of all (%)
	Nov in 2009	Dec in 2009	Jan in 2010	Feb in 2010	Mar in 2010	Apr in 2010	May in 2010	Jun in 2010	Jul in 2010	Aug in 2010	Sep in 2010	Oct in 2010	Sum	
1. <i>Broussonetia</i>	0	0	0	0	0	135	1748	6	1	1	0	0	1891	39.25
2. <i>Humulus</i>	5	0	0	0	0	0	0	0	0	10	440	429	884	18.35
3. <i>Urticaceae</i>	0	0	0	0	0	0	0	0	0	3	215	13	231	4.79
4. <i>Gramineae</i>	6	0	0	1	3	9	37	9	8	8	31	91	203	4.21
5. <i>Platanus</i>	0	0	0	0	0	194	4	0	0	0	0	0	198	4.11
6. <i>Cupressaceae</i>	0	0	2	33	137	4	1	0	0	0	0	0	177	3.67
7. <i>Pinaceae</i>	1	1	0	6	10	46	94	15	1	0	0	1	175	3.63
8. <i>Betulaceae</i>	2	1	1	2	15	35	49	3	0	0	0	0	108	2.24
9. <i>Artemisia</i>	6	3	0	0	1	1	0	0	1	4	39	33	88	1.83
10. <i>Ginkgo biloba</i>	0	0	0	0	0	76	2	1	0	0	0	0	79	1.64
11. <i>Cruciferae</i>	0	1	3	1	1	49	11	0	0	0	0	0	66	1.37
12. <i>Plantaginaceae</i>	39	13	0	0	0	0	0	0	5	4	1	0	62	1.29
13. <i>Palmae</i>	0	0	0	0	0	0	22	32	8	0	0	0	62	1.29
14. <i>Chenopodiaceae/ Amaranthaceae</i>	4	1	0	1	1	0	4	5	4	8	19	11	58	1.20
15. <i>Taxodiaceae</i>	0	0	1	11	24	20	0	0	0	0	0	0	56	1.16
16. <i>Pterocarya</i>	0	0	0	0	1	44	7	1	0	0	0	0	53	1.10
17. <i>Compositae</i>	12	1	0	1	1	0	8	1	0	10	1	13	48	1.00
18. <i>Ulmaceae</i>	0	0	1	4	6	29	0	1	1	0	1	1	44	0.91
19. <i>Moraceae</i>	0	0	1	0	4	11	6	0	1	1	0	0	24	0.50
20. <i>Salix</i>	0	0	0	1	18	3	0	0	0	0	0	0	22	0.46
21. <i>Legume</i>	0	0	0	0	0	0	15	0	0	0	0	0	15	0.31
22. <i>Polygonaceae</i>	0	0	0	0	0	0	0	3	7	1	0	0	11	0.23
23. <i>Apiaceae</i>	0	0	0	0	0	0	7	1	0	0	0	0	8	0.17
24. <i>Liliaceae</i>	7	0	0	0	0	0	0	0	0	0	0	0	7	0.15
25. <i>Rosaceae</i>	0	0	1	0	1	4	0	0	0	0	0	0	6	0.12
26. <i>Buxus</i>	0	0	0	0	0	6	0	0	0	0	0	0	6	0.12
27. <i>Myrtle</i>	0	0	0	0	0	0	1	1	0	0	4	0	6	0.12
28. <i>Papilionaceae</i>	0	0	3	1	0	0	0	0	0	0	0	0	4	0.08
29. <i>Dianthus</i>	0	0	0	0	4	0	0	0	0	0	0	0	4	0.08
30. <i>Populus</i>	0	0	0	0	2	2	0	0	0	0	0	0	4	0.08
31. <i>Ragweed</i>	0	0	0	0	0	0	2	0	0	0	0	0	2	0.04
32. <i>Lamiaceae</i>	0	0	0	1	0	0	0	1	0	0	0	0	2	0.04
33. <i>Ligustrum</i>	1	1	0	0	0	0	0	0	0	0	0	0	2	0.04
34. <i>Rubiaceae</i>	1	0	0	0	0	0	0	0	0	0	0	0	1	0.02
35. <i>Euphorbiaceae</i>	0	0	0	1	0	0	0	0	0	0	0	0	1	0.02
36. <i>Cyperaceae</i>	0	0	0	1	0	0	0	0	0	0	0	0	1	0.02
37. <i>Ailanthus</i>	0	0	0	0	0	0	0	1	0	0	0	0	1	0.02

**Table 2. Monthly distribution of airborne pollen concentrations in the central urban area of Shanghai by the volumetric method with Rotorod Sampler 40 (Continued)**

Pollen species	Pollen concentration(g/m <sup>3</sup> )													Proportion of all (%)
	Nov in 2009	Dec in 2009	Jan in 2010	Feb in 2010	Mar in 2010	Apr in 2010	May in 2010	Jun in 2010	Jul in 2010	Aug in 2010	Sep in 2010	Oct in 2010	Sum	
38. <i>Aceraceae</i>	0	0	0	0	0	0	1	0	0	0	0	0	1	0.02
39. unknown	28	5	4	6	8	10	87	14	6	15	8	16	207	4.30
sum	112	27	17	69	239	678	2106	95	43	65	759	608	4818	100.00

### Pollen species composition

There were obvious differences in the distribution of pollen species throughout the year. Tree pollens were predominantly found in spring. A total of 29 pollen species, amounting to 3023 g/m<sup>3</sup>, were identified from March 1 to May 31, 2010, and constituted 62.74% of the overall pollen count. The most abundant species were *Broussonetia* (62.29%), *Platanus* (6.55%), *Pinaceae* (4.96%), *Cupressaceae* (4.70%) and *Betulaceae* (3.27%). Grass pollens were predominantly found in autumn. A total of 11 pollen species, amounting to 1367 g/m<sup>3</sup>, were identified from September 1 to October 31, 2010, and constituted 28.37% of the total pollen count. The most abundant species were *Humulus* (63.57%), *Urticaceae* (16.68%), *Gramineae* (8.93%), *Artemisia* (5.27%) and *Chenopodiaceae/Amaranthaceae* (2.19%). As shown in **Figure 2B**, grass and tree pollens comprised 35.28% and 60.42% of the overall pollen count respectively, with 4.30% of the pollens unknown.

### Comparison of pollen species and counts between the two methods

As shown in **Tables 1 and 2**, the main pollen species were basically the same using both methods, with 37 and 39 species obtained by the gravitational and volumetric methods, respectively. However, *Urticaceae* was identified only using the volumetric method, and constituted 4.79% of the total pollen count throughout the year, following only *Broussonetia* and *Humulus* in abundance. A total of 32 species were identified by both the gravitational and volumetric methods. Five species were identified only by the gravitational method, including *Paulownia* (1.13%), *Ricinus* (0.23%), *Ericaceae* (0.13%), *Koelreuteria* (0.07%), and *Campanulaceae* (0.04%) in order of abundance. A total of seven species were identified only by the volumetric method, including *Urticaceae* (4.79%), *Polygonaceae* (0.23%), *Apiaceae* (0.17%), *Liliaceae* (0.15%), *Papilionaceae* (0.08%), *Lamiaceae* (0.04%) and *Aceraceae* (0.02%) in order of total pollen proportions.

### Correlation between the two methods for pollen counting

Airborne pollens showed a peak from March to May using both methods, with tree pollens in greatest abundance, and a predominance of *Broussonetia*; there was a small peak from September to October, with grass pollens in greatest abundance, and a predominance of *Humulus*. The total pollen counts detected by the two methods were significantly correlated ( $r=0.622$ ,  $P<0.05$ ) (**Figure 3**).

### Discussion

Rapid economic development, with the consequent changes in lifestyle, eating habits and green vegetation, in addition to the increasingly serious impacts of air pollution which has potentially harmful effects on human health and can promote respiratory diseases, has contributed to the rising prevalence of respiratory allergic diseases. It has also profoundly impacted the quality of life of patients, representing a significant socioeconomic burden. Airborne pollen is a major cause of seasonal allergic rhinitis and asthma.<sup>6,7</sup> Symptom severity is markedly associated with pollen concentration,<sup>12-14</sup> and asthma hospitalization rates increase markedly in the pollen season.<sup>17</sup> Therefore, airborne pollen monitoring is highly important in guiding the clinical management of allergic ailments.

This study showed that airborne pollens are present in the ambient air throughout the year, even in the cold month of January in Shanghai, China. However, distinct seasonal distributions were observed with two peak periods: March to May in spring and September to October in autumn. Tree pollen species were predominant in spring, with *Broussonetia* as the most abundant, not *Cryptomeria* as described in the 1990s.<sup>8</sup> The most abundant pollen groups in autumn were *Humulus*, *Urticaceae* and *Gramineae*. Moreover, *Gramineae* had a biphasic peak in May and September to October. Our study showed that *Broussonetia*, *Pinaceae*, and *Platanus* were predominant in spring, whereas *Moraceae*, *Cinnamomum*, and *Pinaceae* were most abundant in the south-west area of Shanghai with lower amounts of *Broussonetia* and *Platanus*. These regional differences are mainly caused by different plant varieties in various regions. Our samplers were located in the city center, close to the large area of The People's Park green space in West Yan'an Road and Bund Park, close to buildings, shops, roads and residences. *Roussonetia*, *Pinaceae*, and *Platanus* are common greening plants, cultivated as street trees in Shanghai and other cities, and cover a larger area downtown than in the suburbs, leading to its predominance. In a survey of airborne pollens in the southwest area of Shanghai,<sup>9</sup> the location of the sampler was surrounded by rich sources of *Moraceae*, *Pinaceae*, *Cinnamomum* and *Cupressaceae*, resulting in the abundance of these types of pollen.

Compared with studies carried out abroad, the seasonal distributions of airborne pollens were similar, but the pollen species composition showed obvious regional differences. For example, the predominant pollens are *Quercus*, *Morus*, *Pinaceae* and *Platanus* during the spring season in Cincinnati, a city in the northeastern United States.<sup>18</sup> Tree pollens are

predominant in Monterey in Mexico,<sup>19</sup> and account for 72% of the total pollen count, peaking from January to March, of which *Fraxinus* is the most abundant. *Chenopodiaceae*/*Amaranthaceae*, ragweed and pellitory are the main grass pollen species, and are found throughout the year, but are predominant in May, June and December. A 12-year history of local pollen count data showed that tree pollens are the most abundant species in the Washington, DC area, with oak, cedar, and *Moraceae*, as well as *Betulaceae* as the predominant species.<sup>20</sup> In the Malaysian capital of Kuala Lumpur,<sup>21</sup> grass pollen constitutes more than 40% of the total pollen count, and *Gramineae* peaks in the months of March and September, in contrast to our findings. Researchers in Thailand<sup>22</sup> found that grass pollens <40 µm, sedge, *Amaranthus* and fern spores are predominant in Bangkok, Thailand, in a 2012-2013 study excluding an unidentified pollen group; however, there was a shift in pollen peaks to about one to two months earlier than what was observed 35 years before. Ragweed pollen is the most highly allergenic pollen known, and is very important in North America and other countries; it is also the main airborne weed pollen in the city of New York.<sup>23</sup> Ragweed is an exotic invasive species in China, but large-scale dissemination has not been observed domestically.<sup>23,24</sup> The main allergen for hay fever are *Artemisia* in the northern area of China,<sup>23</sup> while *Gramineae* is the main allergenic pollen in the southern area. No ragweed pollens were detected in Shanghai in previous airborne pollen studies.<sup>8,9</sup> Few ragweed pollens were collected in this study, and only in August.

We found that the predominance of some species of pollens (*Broussonetia*, *Humulus*, *Gramineae*, *Pinaceae*, *Platanus*, *Taxodiaceae*, and *Cupressaceae*) was closely related to their individual characteristics, such as total number, size, structure, weight, and presence of sacs or not, among others. The pollens present in larger quantities, smaller volumes or with a lighter weight were more easily collected by the samplers. The three-dimensional structure of a pollen is related to its lift and drag in the air, which affects its floating ability, and impacts on the possibility of being collected. The pollen sac is very important for pollen to spread in the air; it helps pollen (e.g., *Pinaceae*) to float in the air for a long duration and distance. In the actual pollen count analysis, *Broussonetia* was observed large amounts despite its short drift duration. Each *Broussonetia* flower has four anthers; a single male inflorescence has about  $6 \times 10^8$  grains of pollen and about 217 small flowers.<sup>25</sup> *Broussonetia* pollen is very small, i.e. 13~15.8 µm in diameter,<sup>12,26</sup> which allows it to travel easily in the air, with an absolute advantage in total number. In addition, *Pinaceae*, *Platanus*, *Taxodiaceae* and *Cupressaceae* are common greening tree species with wide cultivation areas; *Humulus* and *Gramineae* have the characteristics of a large number of grains with a light weight and small volume; additionally *Pinaceae* pollen has sacs. These properties led them to represent a significant share of the total number of pollens.

As the gravitational method is a high quality approach, inexpensive, easy to use and can monitor the dispersal of airborne pollens in limiting circumstances, it has been used for pollen studies for decades, producing high quantities of

valuable data; moreover, this method is suitable for China's national conditions. There is no doubt that the gravitational method also has disadvantages. The characteristics of pollen and air flow markedly influence the sampling results, since the gravitational method uses passive sampling. In addition, pollens too small to settle on exposure sheets cannot be detected by the gravitational method. However, the gravitational method provides clearer staining with overtly discernible pollen morphology compared with the volumetric method, in which pollen must be identified by a botanist.

By contrast, the Rotorod sampler is designed based on the principle of aerodynamics, and provides 24-hour intermittent sampling and the determination of pollen concentrations in the atmosphere, with an efficiency of more than 80% in collecting pollens and particles greater than 10 µm in diameter. As an active sampling method is used with the Rotorod sampler, sedimentation may affect the results. With particular emphasis on China, counts are easily affected due to poor air quality that can cause even more sedimentation. At present, the volumetric method is used internationally for pollen sampling; thus, it may have more advantages than the gravitational method in scientific research and international exchanges.

Due to the differences in sampling principles and units between the gravitational and volumetric methods regarding the various concepts of pollen content, it is difficult to directly compare both groups of data. In addition, the gravitational method is mainly used domestically, resulting in fewer reports with regard to the correlations between these two methods. This study used both methods simultaneously for the first time to monitor airborne pollen levels in a central urban area of Shanghai, China. As described above, the data obtained using the gravitational method were significantly correlated with those from the volumetric method, and pollen counts collected by both of them were highly correlated. The gravitational method was found to be reliable, providing efficient staining with clearly discernible pollen morphology. It is therefore considered a valuable complement to the volumetric method.

The duration of hay fever mainly depends on the length of the pollination period and how long the allergenic pollen remains in the air. The distribution of airborne pollens in the central area of Shanghai described here constitutes data for future pollen forecasting, so as to provide an important basis for the prevention and treatment of hay fever, while guiding the work of urban greening to some extent and reducing the potential threat of anemophilous pollen-induced diseases. However, pollen amounts are not necessarily related to sensitivity, which will be discussed in the future.

In view of the rich species distribution of airborne pollens in Shanghai and the differences in annual weather, continuous yearly monitoring is necessary to assess the pollen types and concentrations, as well as the law of periodic variation and its relationship with meteorological factors, to help clinicians choose more targeted allergenic reagents for the skin prick test. Patients could also be treated more pertinently, and guided to take protective measures if travelling during the pollen season, or be administered specific immunotherapy.

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## Conflict of interests

All authors declare that they have no any conflicts of interest.

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