Original Article

Preseasonal scattering of *Cryptomeria japonica* pollen in Japan, with reference to the dormancy of the male flowers

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ABSTRACT

Pollinosis caused by *Cryptomeria japonica* is a very serious health problem in Japan. To clarify the relationship between *C. japonica* pollen emissions before the main pollen dispersal season and dormancy of the male flowers, pollen emissions from *C. japonica* trees were observed from November to February, using both flowering tests and an airborne pollen survey. Some *C. japonica* trees studied shed pollen in November and December, by one of two mechanisms. First, some male flowers turned brown and withered, resulting in the formation of small crevices between the scales and pollen leakage. Second, some pollen was released via elongation of the rachis, which is the normal mode of flowering in spring. These early pollen emissions from male flowers were considered to represent a process of self-thinning or unseasonable flowering. *Cryptomeria japonica* pollen counts between October and January were closely related to mean monthly temperatures and airborne pollen counts of the following pollen season. The present study clearly shows that some male flowers of the species can bloom and disperse abundant pollen via blooming or withering in November, if the temperature in October is high, in years of high pollen production.

Key words: *Cryptomeria japonica*, dormancy, male flower, pollen emission, prediction.

INTRODUCTION

Cryptomeria japonica (*C. japonica* D. Don) is one of the most important coniferous species in Japan. Its natural distribution spans the Japanese archipelago, except for Hokkaido (the northern-most island) and Okinawa, the southern-most island. This species now covers 4.5×10^5 ha, accounting for 45% of the afforested area in Japan.[1] *Cryptomeria japonica* disperses abundant pollen from the end of February to early May.[2] Horiguchi and Saito[3] first described allergies to *C. japonica* pollen in 1964. These allergies have since increased rapidly and now affect approximately 10% of the Japanese population.[4] Thus, forecasting pollen emissions from the *C. japonica* forest can play an important role in helping allergen avoidance. The florescence of *C. japonica* male flowers is inhibited by dormancy and is initiated during their rest-breaking phase. Therefore, knowledge of the dormancy characteristics of these flowers is required for precise forecasts of pollen emission. However, there have been few studies of the relationship between pollen dispersal and the dormancy and rest-breaking kinetics of the male flowers.[5] Thus, the present study was undertaken to clarify the relationship between pollen emissions, dormancy and temperature. It was found that male flowers dispersed abundant pollen in the autumn just after maturation in 1994. In order to evaluate the dormancy of *C. japonica* male flowers and pollen release, the authors directly observed pollen emissions from *C. japonica* trees and performed flowering tests every month from December through to February. The relationship between dormancy of the male flowers and temperature during the autumn and winter was also studied.

METHODS

Study site and flowering tests

The study site was established at the Toyama forest experimental station in Toyama Prefecture (36°36′ N 137°40′ E), Japan. Forty-seven *C. japonica* trees bearing abundant male flowers were chosen from a 40-year-old stand for examination. Every 10 days between 16 November 1994 and 12 February 1995, male flowers on each *C. japonica* tree were tapped with a bamboo stick to assess whether pollen was being shed.

A sample twig with male flowers was collected from each of the 47 *C. japonica* trees once a month between 16 November 1994 and 6 March 1995. Each of these twigs was cut to a length of 30 cm and was placed in a glass tube (2.5 cm diameter; 12 cm long), which was filled with water.[5] The water was changed daily. Tubes were placed in an incubator at 10°C, where the light intensity was maintained at 20 000 lux for 20 h/day. Each twig was examined daily for pollen emission until all the male flowers on the twig had released pollen. Pollen emission from the male flowers was confirmed by tapping them with a short stick over a black laboratory table and the developmental status of the flowers was observed under a stereoscopic microscope.

Airborne pollen survey

An airborne pollen survey was conducted using a Durham's sampler[6] on the roof of the Toyama Medical and Pharmaceutical University in Toyama Prefecture (36°40′ N, 137°07′ E) between May 1988 and April 1997. A glass slide smeared with petrolatum was collected each day and the pollen on the glass slide was dyed with methyl violet. The number of pollen grains on the glass slide was counted under a microscope at ×200 magnification[7] and daily airborne pollen contents were expressed as counts/cm^2. The relationships between monthly airborne pollen counts in October, November, December and January, the total airborne pollen counts the following season and weather factors were analyzed. For statistical evaluation, both multiple linear regression analysis and analysis of variance (ANOVA) were used.

RESULTS

Thirteen of 47 *C. japonica* trees studied shed pollen on 16 November 1994 when male flowers on the trees were tapped with a bamboo stick (Fig. 1), decreasing to eight
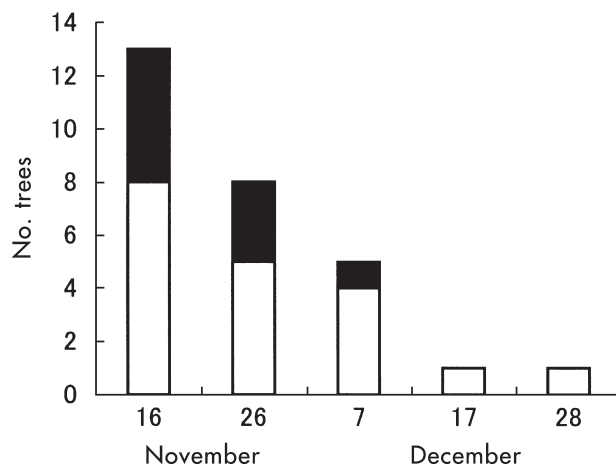

Fig. 1 Number of *Cryptomeria japonica* trees from which macroscopic pollen was released following tapping of the male flowers with a bamboo stick. (□), pollen released from trees with male flowers in bloom; (■), pollen released from trees with withered flowers.
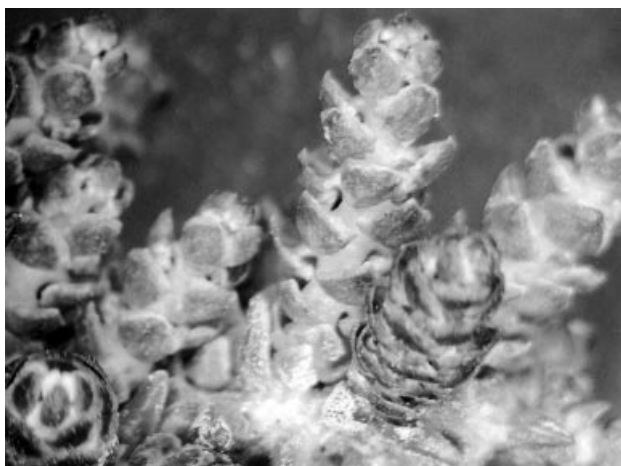

Fig. 2 Representative male flower dispersing pollen via elongation of the rachis.

and five trees on 26 November and 7 December, respectively. On both 17 December and 28 December, only one tree shed pollen and none of the trees released pollen after 28 December. We observed two mechanisms whereby pollen was shed from male flowers. The first was the way in which pollen is shed in the spring; that is, the rachis elongated, gaps developed between the scales and pollen was dispersed between the gaps (Fig. 2). The second mechanism of shedding pollen involved the changing of the color of the male flowers to brown and withering of the brown male flowers, which caused small
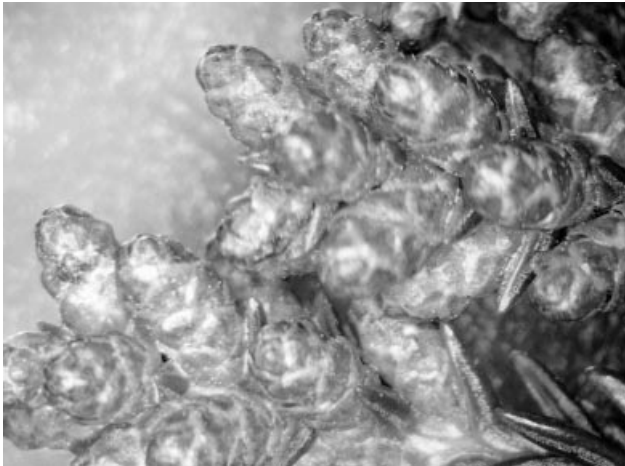
Fig. 3   Representative withered male flowers dispersing pollen.

Fig. 6   Total airborne Cryptomeria japonica pollen counts in October (▨), November (▧), December (■) and January (□) between 1988 and 1998.
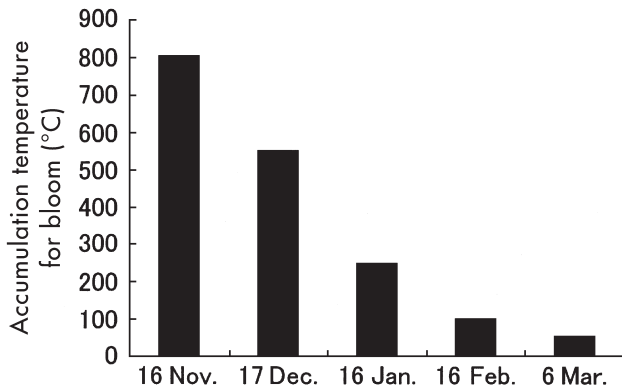

Fig. 4   Mean accumulation temperature needed to induce flowering of all male flowers on twigs obtained on the dates indicated (incubation temperature 10°C).

Fig. 7   Relationship between total airborne pollen counts in October, November, December and January and airborne pollen counts of the following season (y = 0.0092x − 7.2585; R^2 = 0.9071).
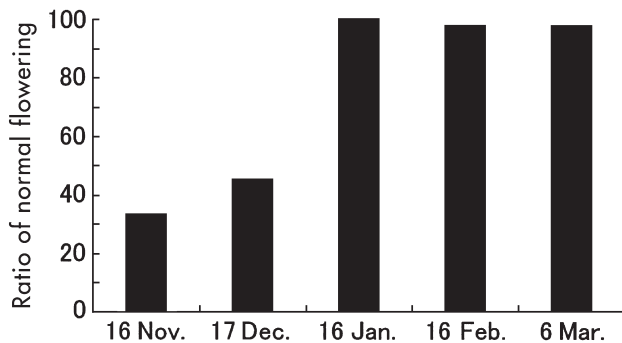

Fig. 5   Percentage of male flowers incubated at 10°C that released pollen via rachis elongation.

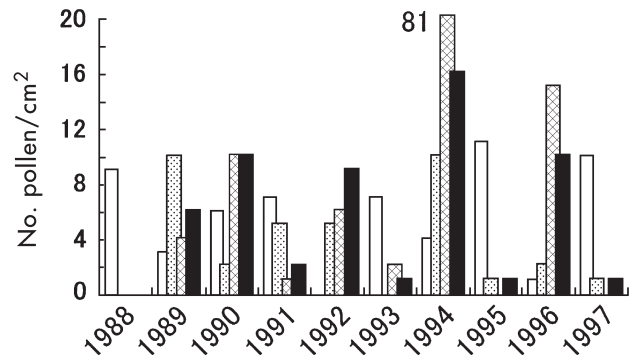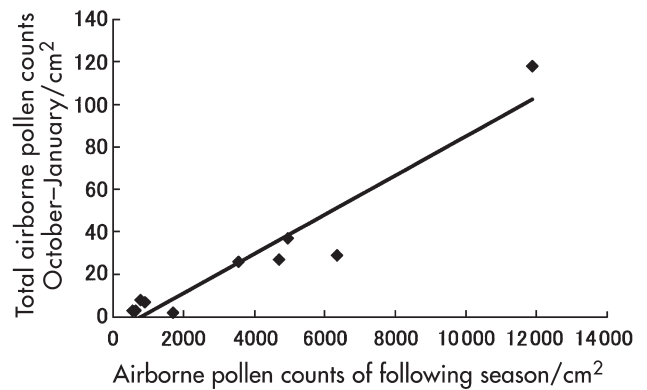crevices to form between the scales and, thus, allowed pollen to leak out (Fig. 3). On 16 November, 26 November and 7 December, we observed male flowers on eight, five and four trees, respectively, releasing pollen by elongation of the rachis. On the same dates, five, three and one tree, respectively, were shedding pollen from withered male flowers. On 17 December and 28 December, only one C. *japonica* tree released pollen, on both occasions by elongation of the rachis. The number of male flowers shedding pollen by the rachis elongation and flower withering mechanisms during the autumn comprised approximately 10 and 30% of all male flowers on the twigs, respectively. The remaining male flowers on the twigs bloomed normally in the flowering season.

All male flowers incubated at 10°C eventually released pollen via elongation of the rachis or withering. The mean accumulated temperature required to induce all

Table 1   Results of multiple correlation analysis of the mean monthly temperature, the pollen count of that month and the pollen count of the following season between 1988 and 1997

Month	Multiple *R*	*F*	Sig. *F*
October	0.729	3.4	0.13
November	0.933	20.285	0.02
December	0.975	56.982	0
January	0.89	11.422	0.09

Table 2   Mean monthly temperature of October, November and December between 1988 and 1997 and average temperature of October, November and December over the past 30 years

	Mean temperature (°C)		
	October	November	December
1988	14.8	8.0	4.8
1989	15.1	11.6	5.8
1990	16.4	12.3	7.1
1991	16.5	10.3	6.9
1992	16.2	11.1	6.1
1993	15.2	11.7	5.6
1994	18.0	11.7	6.0
1995	17.4	8.9	4.0
1996	16.1	10.4	6.1
1997	15.5	11.6	6.5
Average	15.6	10.2	5.2

Average; average temperature at Toyama Meteorological Observatory in October, November and December over the past 30 years.

male flowers on the twigs to release pollen by elongation of the flower or withering differed according to the month in which the twigs were picked. The mean accumulated temperature required was extremely high in November, but it gradually decreased in successive months through to March. The mean cumulative temperature requirement for male flowers on twigs picked on 17 November was 804°C, while the corresponding temperature sum for the male flowers on twigs picked on 6 March was just 85°C (Fig. 4). Little pollen was released via elongation of the rachis on either 16 November or 17 December, but the abundance greatly increased after 16 January (Fig. 5).

The monthly airborne pollen count between October and January varied between years, ranging between 3 and 118, but the monthly airborne pollen counts in the period October 1993 to January 1994 were extremely high. Generally, in the period 1988–98, the monthly airborne pollen counts tended to be low in October and high in November (Fig. 6). The airborne pollen counts during the pollen season varied widely between 1989 and 1997, ranging between 557 and 11 895. There was a high positive correlation ($R^2 = 0.91$) between total airborne pollen counts in October, November, December and January and the airborne pollen counts in the following season (Fig. 7). Multiple correlation analysis of the mean temperature in October, the airborne pollen counts for that month and the pollen count in the following season detected no significant relationship at a 5% probability level. However, similar analysis indicated that there were significant relationships ($P < 0.05$) among the monthly mean temperatures in November, December and January, the airborne pollen counts of the respective months and the pollen counts in the following season (Table 1).

DISCUSSION

Cryptomeria japonica is a monoecious plant. Most male flower buds of the species are formed from late July to late August and they continue to develop gradually until the middle of October.[8] In early September, most pollen mother cells have reached the tetrad stage and they finally reach the mature stage with two nuclei by mid-October.[9,10] Male flowers generally bloom, via elongation of the rachis, from February to April in the following year. Airborne pollen counts in 1995 were the highest for 16 years according to a long-term airborne pollen survey.[11] Thus, many male flowers were produced in 1994.

In the present study, it was observed that pollen was shed from the male flowers by two mechanisms. In the first, male flowers simply withered. In these cases, the pollen emerged between small crevices in the scales and there was no elongation of the rachis. In the male flower, 18–25 sporangia are spirally arranged[12] and they readily dehisce following a minor shock, such as a light tap. Consequently, when the sporangia dehisce, abundant pollen can leak out between the scales even without elongation of the rachis. Thus, this mechanism of pollen dispersal cannot be regarded as florescence of the male flowers. The number of trees that shed pollen via the male flowers withering declined sharply as the autumn progressed and no trees shed pollen by this mechanism from 7 November onwards. This phenomenon is commonly observed in years with abundant pollen.[13] Similarly, *Diospyros kaki* trees drop some of their fruit within a few months of their initiation when many fruits are produced.[14] The withering of male flowers is considered to be a kind of physiologic dropping trait, which occurs

most strongly when many male flowers develop. The other means of pollen dispersal observed in the present study was the standard mechanism, involving elongation of the rachis, generally seen in the spring. Male flowers on eight of 47 C. *japonica* trees studied were releasing pollen via elongation of the rachis on 16 November. However, this number fell as the temperature cooled and no trees released pollen via elongation of the male flowers after 28 December.

In woody perennials, it is known that under conditions of long days and moderate temperatures, resting buds will eventually begin to grow, even if they have not been sufficiently chilled after vegetative maturation to induce general flushing.[15] Similarly, in annual and biennial species, plants eventually flower at most non-vernalizing temperatures.[16] The phenomenon of flowers blooming before the dormancy period has also been observed in species of plum, apple, peach and pear when they have been exposed to drought or strong wind.[17] The mean temperature in October 1994 was 18.0°C, 2.4°C higher than the mean October temperature over the past 30 years (Table 2), but the monthly precipitation figures for October and November differed little from the long-term averages. Therefore, it is suggested that the high temperature in October 1994 induced florescence in some male flowers when they were reaching maturation.

Fuchigumi and Nee[15] developed a numerical concept, the degree of growth stage (°GS) model, to quantify the annual development of temperate zone woody species. Applying their model to the development of C. *japonica* male flowers, the cumulative temperature required for male flowers to bloom should be lowest just after maturation of the flowers and it should then increase. However, in the present study, the highest accumulated temperature required for all male flowers to disperse pollen was found on 16 November, just after the male flowers had matured. The accumulated temperature needed for anthesis of the male flowers then decreased in successive months through to March.

Of all the male flowers that shed pollen, only a small percentage on twigs obtained in November bloomed via rachis elongation. Of the twigs obtained in December, there was a slight increase in the percentage of male flowers that bloomed in this way and most male flowers on twigs obtained after January bloomed via this mechanism. This suggests that sufficient chilling generally occurs in winter to allow the male flowers to bloom normally. It has been shown that azalea plants can flower early if they are chilled just after maturation of the pollen and ovule.[18]

This species can bloom any time after November in natural conditions. Therefore, the chilling requirements of C. *japonica* male flowers are similar to those of azalea and they differ in this feature of bud dormancy to most temperate zone woody species.

The present study clearly shows that some male flowers of the species can bloom and disperse abundant pollen via blooming or withering in November, if the temperature in October is high, in years with high pollen production. There was a significant positive relationship ($P < 0.05$) between the monthly mean temperatures in November, December and January, airborne pollen counts of the respective month and pollen counts in the following season. This information should be useful for forecasting airborne pollen abundance and, thus, help avoid C. *japonica* pollinosis.

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