

TIME OF DAY OF POLLEN SHEDDING OF SOME HAY FEVER PLANTS

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POLLEN shedding and dispersal is of particular scientific interest to the botanist and the plant breeder in their consideration of the phenomena of pollination, fertilization, and seed setting. But this subject is also of interest to the medical profession in the treatment of asthma and pollinosis.

The present study grew out of a plant breeding investigation aimed at determining the time of day when grasses shed pollen. This information is needed by plant breeders as a basis for collecting large quantities of viable pollen and for making hybridizations in the field at the time when there is the least danger of contamination by foreign pollen in the air.

The same information might be useful to the medical profession in the collection of large quantities of pollen for use in clinical medicine.

Surface reactions caused by pollen, such as hay fever, asthma, and dermatitis of the exposed parts, vary in intensity at different seasons, during different weeks of the same season, and at different times during a given day.² Numerous kinds of pollen may be present in the air at once, but usually only a few of them will be of any significance as hay fever agents.¹¹

Numerous reports are available in the literature relative to the time of day certain species shed pollen. However, most of these reports are based on observation rather than experimental measurement of the amount of pollen liberated hourly from plants during the day.

Knuth¹⁰ summarizes the observations of numerous workers in his monograph "Handbook of Flower Pollination." In general the reports of time of day of pollination are in fair agreement, but numerous exceptions can be found where investigators report entirely different times of the day for pollen shedding for a species in the same region.

Jensen and Bøgh⁷ made counts of the number of pollen grains caught on petroleum jelly-coated microscope slides which were exposed at various intervals during the day. They found this scheme of determining time of day of pollen shedding to be very accurate.

In a study of 29 grasses at Lincoln, Neb., in 1944 and 1945, Jones and Newell⁸ used a similar technique and found that most of the grasses required 2 to 6 hours in each of several successive days for the completion of pollen shedding. When meteorologic conditions were optimum for blooming, these daily pollination periods occurred rather regularly at the same time of day for any specific grass. Before or after this short period of active shedding, little pollen was present in the air, even in the center of the grass field. Daily pollen shedding occurred partially or entirely during periods of darkness for thirteen of the grasses studied. Others normally shed pollen in daylight.

The assumption that the presence of excitants is a common characteristic of all grass pollen seems warranted from numerous tests.¹ In general, the

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self-fertilized grasses, such as wheat, barely, and oats, and the grasses (including corn) which produce the larger and less buoyant pollen, are not considered of great importance as excitants. The relatively small, buoyant pollen grains of widely distributed grasses, such as Kentucky bluegrass, Timothy, and Bermuda grass, have been listed as the most important in contributing to the hay fever malady.

Geisler,⁶ described the general characteristics of grass pollen and presented drawings of pollen from 32 different grasses obtained from mounted herbarium specimens. She studied grasses from four habitats, viz., aquatic, wet lowland, mesophytic, and prairie. She reports that the pollen of prairie grasses is markedly larger than that of more moist habitats with the exception of the pollen of *Zizania aquatica*, which is similar in size to the pollen of andropogon and Sorghastrum. This fact made it impossible to separate prairie from aquatic grasses on the basis of pollen size differences.

Results of studying pollen from the same grass stand in two different years indicate that pollen size may vary as a result of environmental conditions.⁹ Seventeen of 31 grasses produced pollen with no significant size variation between the two years, 5 showed significant differences at the 5 per cent level and 8 showed significant differences at the 1 per cent level. Thus seasonal environment has a pronounced effect on the pollen size of some grasses.

In a recent series of articles, Durham⁷⁻⁹ reported the specific gravity, rate of fall, and size of pollen of numerous species of plants causing hay fever.

METHODS

Forty-five species of plants were studied at Lincoln, Neb., in 1945 and at Stillwater, Okla., in 1946 and 1947, to determine pollen size and the time of day and time of year that pollen shedding occurred. The species studied are given in Table I.

Pure samples of pollen were collected from each plant and stored in glycerine-alcohol solution. Measurements of pollen diameters were made from permanently mounted slides according to the method described by Jones and Newell.⁹

To determine the time of day a species sheds its pollen, four microscope slides which had an area of one square inch coated with a thin film of petroleum jelly were exposed at elevations near the height of the plant in the center of the selected area. The slides were placed in holders that were attached to weather vanes at an angle of 45° (Fig. 1). This method of exposure kept the petroleum jelly-coated side of the slide exposed against the prevailing wind and facilitated catching the pollen that was impinging against the slide, whether it was blown through the air or fell as a result of gravity. The total number of pollen grains on 10 random low-power microscope fields (16 sq. mm. area) per slide was used as a measure of the amount of pollen shed.

Close observation of the anthers previous to blooming made it possible to predict when pollen shedding might be initiated by a given species. For these

studies, preliminary exposures were made every five to six hours during the first days when the investigation of a species was begun. After the general time of day when pollen was being shed from the anthers was determined, intensive studies were continued for two to six days. Exposures were made at 30 minute intervals during the period of active pollen shedding. When microscope examination revealed no pollen was being caught on the slides, the 30 minute time exposures were discontinued. Slides were left exposed until the following day, so that counts of pollen could be made for each 24 hour period, as well as for periods of several days. The counts made at 30 minute intervals were recorded on the basis of central standard time.

Aids used in identifying pollen included size and shape measurements, knowledge of which kinds of plants were shedding pollen at the same time of year, the time of day that each plant sheds its pollen, and the mass effect of the pollen under study. Permanent mounts of different pollens were also used for comparison.



Fig. 1.—A weather vane with microscope slide holder attached, as used in this study. Grass plants in background.

EXPERIMENTAL RESULTS

Shape, Color, and Size of Pollen.—The pollen of grasses is characterized by a somewhat spherical shape and yellowish color. Its form varies from the ellipsoidal shape of rye pollen to the almost spherical shape representative of

TABLE I. CLASSIFICATION OF 45 SPECIES ACCORDING TO DIAMETERS OF POLLEN GRAINS AND TIME OF DAY AND SEASON OF POLLEN SHEDDING FROM MEASUREMENTS AND OBSERVATIONS MADE IN THE SEASONS OF 1945 AT LINCOLN, NEBRASKA, AND 1946 AND 1947 AT STILLWATER, OKLAHOMA.

BOTANICAL NAME	COMMON NAME	MEAN DIAMETERS AND STANDARD ERRORS IN μ^*	TIME OF DAY OF POLLEN SHEDDING	GENERAL SEASON OF POLLEN SHEDDING
<i>I. Diameters of 20 to 30 μ</i>				
<i>Alopecurus carolinianus</i>	Swamp foxtail	28.4 $\pm$ 0.1	5-8 P.M.	May, June
<i>Ambrosia trifida</i> †	Giant ragweed	20.1 $\pm$ 0.2	7 A.M.-1 P.M.	Aug., Sept., Oct.
<i>Ambrosia psilostachya</i> †	Western ragweed	25.0 $\pm$ 0.4	7 A.M.-1 P.M.	June, July
<i>Calamagrostis epigeios</i>	Sand lovegrass	27.3 $\pm$ 0.6	7-11 A.M.	Aug., Sept.
<i>Eragrostis trichodes</i>	Common reed	26.3 $\pm$ 0.4	6-10 A.M.	Aug., Sept.
<i>Phragmites communis</i>	Kentucky bluegrass	26.4 $\pm$ 0.4	5-8 A.M.	May, June
<i>Poa pratensis</i>		28.4 $\pm$ 0.9		
<i>II. Diameters of 30 to 40 μ</i>				
<i>Andropogon ischaemum</i>	Yellow bluestem	33.6 $\pm$ 0.8		Aug., Sept.
<i>Andropogon saccharoides</i>	Silver beardgrass	34.2 $\pm$ 0.7		Aug., Sept.
<i>Arrhenatherum elatius</i>	Tall oatgrass	37.6 $\pm$ 0.4	3-7 P.M.	May, June
<i>Bouteloua curtipendula</i> †	Side-oats grama	35.7 $\pm$ 0.5	4-9 A.M.	July, Aug., Sept.
<i>Bouteloua gracilis</i>	Blue grama	30.8 $\pm$ 0.6	3-9 A.M.	July, Aug., Sept.
<i>Buchloe dactyloides</i>	Buffalograss	37.4 $\pm$ 0.7	6-11 A.M.	May, June
<i>Dactylis glomerata</i>	Orchardgrass	36.2 $\pm$ 0.3	4-10 A.M.	June, July
<i>Festuca elatior</i> †	Meadow fescue	37.4 $\pm$ 0.7	3-8 P.M.	June, July
<i>Festuca ovina</i>	Sheep fescue	35.4 $\pm$ 0.7	4-8 P.M.	May, June
<i>Koeleria scoparia</i>	Mexican firebush	33.1 $\pm$ 0.5	7 A.M.-4 P.M.	July, Aug., Sept.
<i>Koeleria cristata</i>	June grass		3-7 P.M.	May, June
<i>Plalaris arundinacea</i>	Reed canarygrass	37.5 $\pm$ 1.2		June, July
<i>Phleum pratense</i>	Timothy	36.6 $\pm$ 0.9	4-9 A.M.	June, July
<i>Stipa viridula</i>	Green needlegrass	35.0 $\pm$ 0.6		May, June

III. Diameters of 35 to 50 μ

Andropogon furcatus	Big bluestem	40.2 $\pm$ 0.6	4-7 A.M.	Aug., Sept.
Andropogon hallii	Sandhill bluestem	40.2 $\pm$ 0.9	4-9 A.M.	Aug., Sept.
Agropyron cristatum	Crested wheatgrass	43.0 $\pm$ 0.6	2-6 P.M.	May, June
Agropyron elongatum	Long-spiked wheatgrass	45.5 $\pm$ 0.7	2-6 P.M.	June, July
Agropyron intermedium	Intermediate wheatgrass	49.1 $\pm$ 1.3	2-6 P.M.	June, July
Agropyron repens	Quackgrass	45.1 $\pm$ 0.6	2-6 P.M.	June, July
Agropyron smithii	Western wheatgrass	47.5 $\pm$ 0.6	2-6 P.M.	June, July
Bromus inermis	Brome grass	42.1 $\pm$ 0.7	2-7 P.M.	May, June
Elymus junceus	Russian wildrye	43.7 $\pm$ 0.9	3-7 P.M.	May, June
Elymus virginicus	Virginia wildrye	43.6 $\pm$ 0.6		June, July
Hordeum jubatum	Squirreltail	43.9 $\pm$ 0.1		May, June
Hordeum pusillum	Little barley	38.7 $\pm$ 0.8		May, June
Panicum virgatum	Switchgrass	39.2 $\pm$ 0.8	10 A.M.-2 P.M.	July, Aug., Sept.
Sorghastrum nutans	Indiangrass	44.5 $\pm$ 0.6	6-10 A.M.	Aug., Sept.
Sorghum vulgare	Sorghum	45.4 $\pm$ 0.8	4-10 A.M.	Aug., Sept.
Sorghum vulgare sudanense	Sudan	48.1 $\pm$ 0.7	6-12 A.M.	Aug., Sept.
Spartina pectinata	Sloughgrass	37.3 $\pm$ 0.7	4-10 A.M.	Aug., Sept.
Tripsacum dactyloides	Gamagrass	43.7 $\pm$ 0.5		July, Aug.

IV. Diameters of 45 to 65 μ

Avena sativa	White oat	50.6 $\pm$ 0.9		May, June
Secale cereale	Balto rye	54.4 $\pm$ 1.4	3-11 A.M.	May, June
Triticum vulgare	Pawnee hard wheat	56.7 $\pm$ 0.8	4-10 A.M.	May, June

V. Diameters of 80 to 100 μ

Zea mays†	Yellow dent corn	93.0 $\pm$ 1.1	7 A.M.-2 P.M.	July, Aug.
Zea mays	Popcorn	99.5 $\pm$ 0.8	7 A.M.-2 P.M.	July, Aug.
Zea mays†	Sweet corn	94.2 $\pm$ 0.5	7 A.M.-2 P.M.	July, Aug.

*All measurements made on grains expanded in glycerine-alcohol solutions according to method described in Reference 2.

†Studied at both Lincoln, Neb., and Stillwater, Okla.

‡Studied only at Stillwater, Okla.

most of the grasses studied. The color of the pollen varies from a deep yellow in some varieties of corn to an almost colorless condition in swamp foxtail, with the pollen of most grasses having a creamy to yellowish color. Because of the similarity of the various kinds of grass pollen, neither shape nor natural color appear to be of value in identifying pollen of the different grass species causing hay fever.

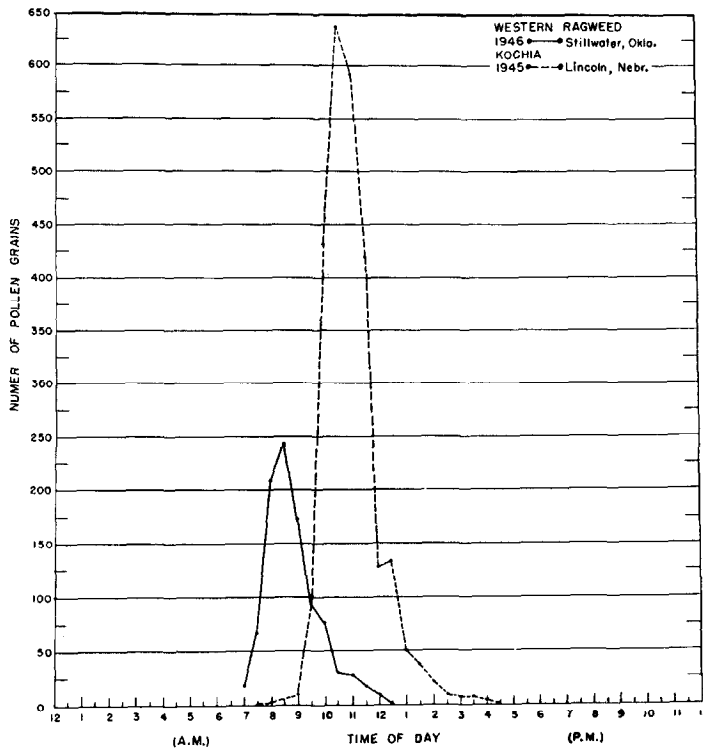


Fig. 2.—Time of day of pollen shedding of Kochia at Lincoln, Nebraska during 1945, and of western ragweed at Stillwater, Oklahoma, 1946.

Diameter measurements of pollen were made in an effort to identify and to determine buoyancy of pollen of different species. The mean diameters of pollen varied from $20.1 \pm 0.2 \mu$ for giant ragweed to $99.5 \pm 0.8 \mu$ for popcorn with 32 of the 45 species having pollen grains between 30 and 50μ (Table I).. Diameter of pollen is related to size and rate of fall.⁵ Durham^{3, 4} gives the specific gravity and experimental rate of fall when compared to short ragweed of eight species which were studied in the present investigation. According to his results and the size measurements made in this study, the small pollen grains of low specific gravity, such as giant ragweed and western ragweed fall more slowly than the large heavy pollen of rye and corn. Mexican firebush, Kentucky bluegrass, Orchard grass, and Timothy were intermediate in rate of fall.⁴

Time of Day of Pollen Shedding.—It should be emphasized that observation alone is not always completely reliable to determine the time of day and

the number of days that species shed pollen. With some plants which are not prolific pollen producers, the exposure of slides has proved the inadequacy of casual observation.

Thirty-five species were studied to determine the time of day they shed pollen. The data are summarized in Table I; however, six species, which were considered representative of the entire group, are discussed in detail. These species include Mexican firebush, smooth bromegrass, giant ragweed, western ragweed, sweet corn, and rye.

Mexican firebush: Kochia or firebush is an escaped ornamental that is becoming a serious hay fever and weed pest in the north central United States. It produces large quantities of pollen from July 10 to September 10 at Lincoln, Nebraska, and is prevalent in nearly all fields and waste places.

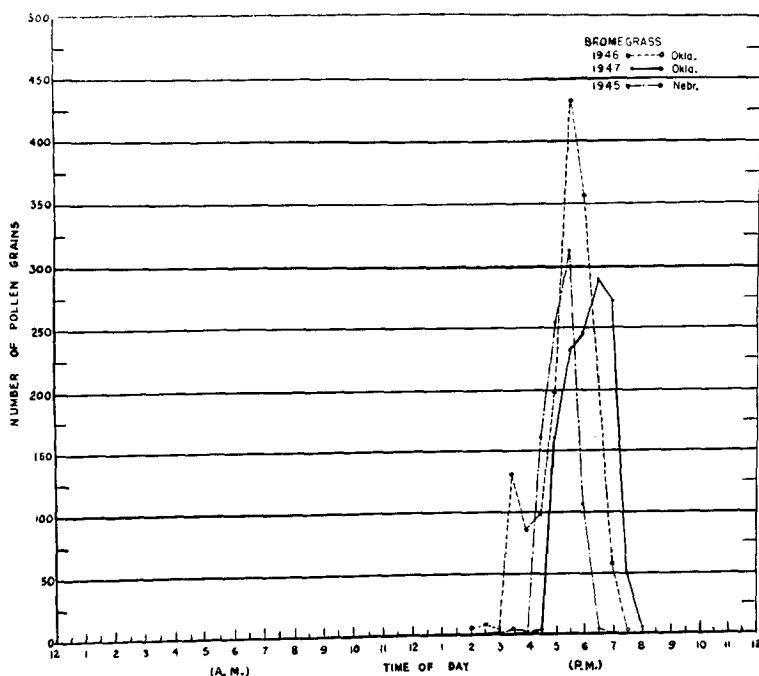


Fig. 3.—Time of day of pollen shedding of bromegrass at Lincoln, Nebraska in 1945 and at Stillwater, Oklahoma in 1946 and 1947.

The seriousness of Kochia as a hay fever plant was brought to the attention of the author by a colleague. He made trips daily at about 10:00 A.M. to experimental plots in the field. Each day during pollen season he would develop severe symptoms of hay fever following his field visits. A study made in 1945 at Lincoln, Nebraska, showed that Kochia shed 94 per cent of its average daily total pollen between 9:00 A.M. and 12:30 P.M., with the peak of shedding occurring at 10:30 A.M. (Fig. 2). Very little pollen was caught on exposed slides after 2:00 P.M. On the basis of these data, my colleague was advised to visit the experimental plots after 2:00 P.M. each day. This change in his schedule resulted in his having very light symptoms of hay fever.

*Smooth brome*grass: The dispersal of smooth brome grass pollen was studied for six days at Lincoln in 1945. On the average, it required only 15 minutes after blooming until the anthers started dehiscing and shedding pollen. Pollen grains of brome grass are only $42.1 \pm 0.7 \mu$ in diameter and are very buoyant. In a few minutes after maximum blooming, "clouds" of yellow pollen were observed as breezes swept across the field. These observations were substantiated by data obtained from slides exposed for 30 minute periods. Pollen was caught on the slides from 2:30 to 7:00 P.M., but 99.5 per cent of the average daily total was shed between 4:00 and 6:30 P.M. (Fig. 3).

Temperatures of 80° to 85° F. were found to be optimum for the blooming of this grass; however, active blooming has been observed at temperatures between 74° and 90° F. Temperatures lower than 70° F. inhibited blooming on June 14 and 15, 1945.

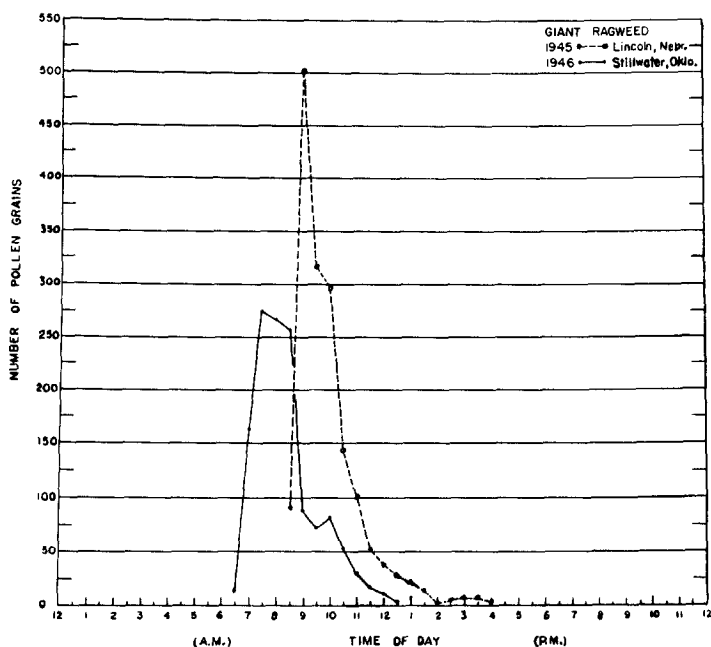


Fig. 4.—Time of day of pollen shedding of giant ragweed at Lincoln, Nebraska, in 1945 and at Stillwater, Oklahoma in 1946.

Records of brome grass pollination at Lincoln for the period 1936 through 1945 show the seasonal pollen shedding occurring in late May or early June. The pollen season lasted only for 11 days in 1944 and for 14 days in 1945. Brome grass produces a tremendous amount of pollen and is grown on a large acreage, and therefore, is a hay fever problem for a short period each season. However, since its daily pollen shedding period is confined to a short period in late afternoon, it is possible to avoid exposure to the air-borne pollen.

Slide exposures were made on four days in the second and third weeks of May in 1946, and during early June in 1947, for the investigation of daily pollen shedding of brome grass at Stillwater, Oklahoma. The majority of pollen in both years was caught during the late afternoon and early evening (Fig. 3).

The days on which studies were made in 1947 were warmer than those in 1946. Clear skies, low relative humidity, and strong winds were common. The temperatures between noon and 4:00 P.M. increased each successive day from June 4 to June 9, inclusive. Likewise, the time of maximum daily pollen shedding each successive day was consistently later by approximately 30 minutes. Apparently the delay in maximum shedding on successive days was due to rather complete blooming and shedding each day; thus, a little more time was required for growth and maturation of the anthers for the next day's pollen supply. Even though optimum temperatures for blooming and shedding of approximately 86° F. prevailed near noon each day, no shedding occurred at that time. Apparently the duration of light and heat had not been sufficient by then to mature the anthers. The time of day of pollen shedding was found to be a little later at Stillwater, in 1946 and 1947 than at Lincoln in 1945.

Giant ragweed: Giant ragweed is one of the heaviest producers of pollen of any species known to cause hay fever. It is fortunate that this species is restricted to certain soil sites and thus is not widespread in most regions. Pollen shedding was studied on Aug. 24 and 25, 1945 at Lincoln. Eighty-eight per cent of the average daily total was shed between 8:00 and 11:00 A.M. (Fig. 4).

Studies were conducted at Stillwater on Sept. 5, 6, 9, and 11, 1946. Pollen shedding was initiated earlier in the day than at Lincoln. About 95 per cent of the average daily total of pollen was shed between 7:00 and 10:30 A.M.

The pollen season at Lincoln in 1945 extended from August 4 to September 30, with the peak occurring between August 24 and September 3. At Stillwater in 1946, the pollen season lasted from August 1 to October 10, with the peak between September 14 and 24. The principal effect of latitude appeared to be on dates rather than on length or severity of the pollen season.

Rainfall on the morning of September 10 delayed pollen shedding for approximately two hours. Apparently the amount of pollen shed on September 10 was only slightly less than normal.

Western ragweed: Western ragweed is a perennial of widespread distribution in the Corn Belt and Great Plains. It normally produces a large daily quantity of pollen from July 1 to September 15, with the peak of production occurring about August 15 to 30 at Stillwater.

As an average for three days in September 1946, pollen shedding extended from 6:30 A.M. to 12:30 P.M., with the period of maximum shedding at 8:30 A.M. Approximately 90 per cent of the average daily pollen supply was shed between 7:00 and 10:00 A.M. (Fig. 2).

Rain delayed pollen shedding for two hours on September 10. After the relative humidity returned to normal, tremendous quantities of pollen were shed about 10:30 A.M.

Sweet corn: The first few pollen grains were caught between 6:30 and 7:00 A.M. on slides exposed in the center of a field of sweet corn for four days in late June and early July in 1946. The rate of pollen shedding increased rapidly until the maximum was reached between 7:30 and 8:00 A.M. (Fig. 5).

Slide studies were made on June 23, 24, and 25 in 1947. Meteorological conditions and the daily pollination cycles were very similar on each of these days. Heavy shedding extended from 8:00 to 11:00 A.M., with the peak of pollen shedding occurring between 8:30 and 9:00 A.M.

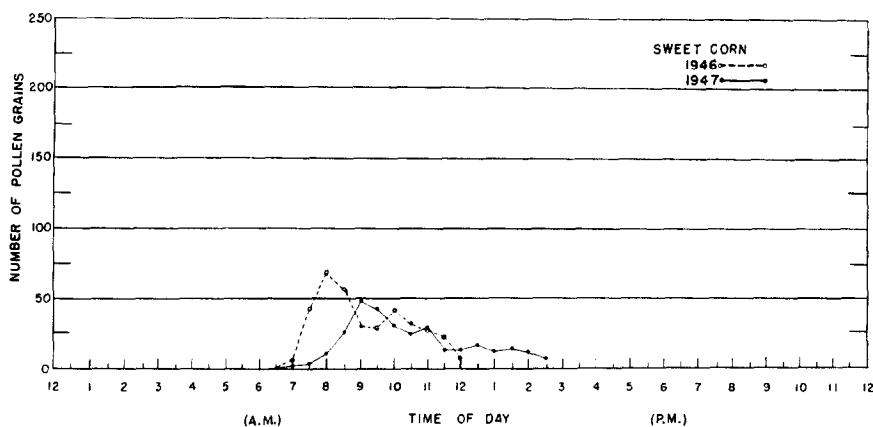


Fig. 5.—Time of day of pollen shedding of sweet corn at Stillwater, Oklahoma in 1946 and 1947.

The maximum rate of pollen shedding occurred an hour earlier in 1946 than in 1947, apparently because of lower relative humidities and higher temperatures in the early morning on the days of study in 1946.

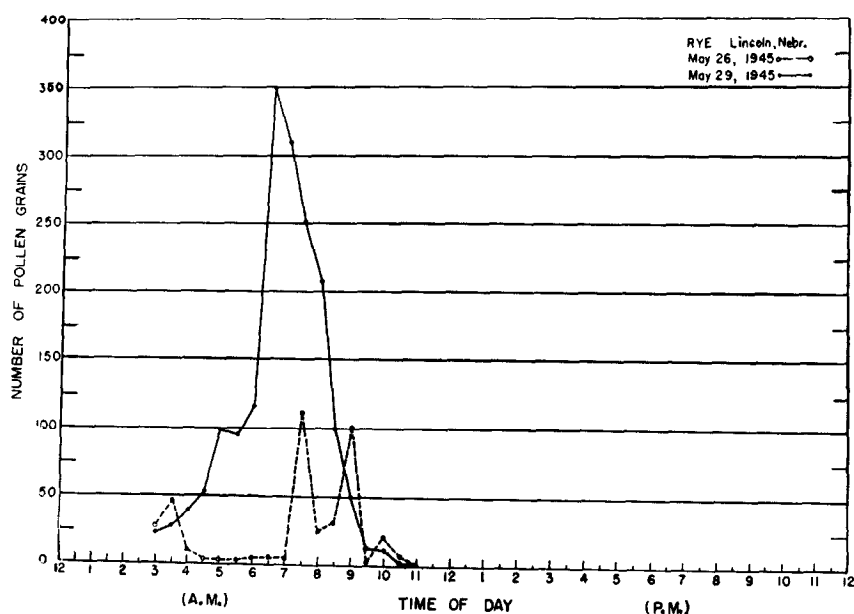


Fig. 6.—Time of day of pollen shedding of rye at Lincoln, Nebraska in 1945.

Sweet corn pollen was found to be $94.2 \pm 0.5 \mu$ in diameter in 1947 and appeared to be the least buoyant of any grass pollen. Even though sweet corn

and field corn are planted on large acreages, the number of plants per acre is small and they have not proved to be serious hay fever pests. The daily pollen production was shed in a short period (Fig. 5).

Rye: The effect of wind on the dispersion of rye pollen was studied on May 26 and 29, 1945. It was necessary to begin exposures of slides before dawn to determine the extent of pollen shedding. Slides were placed on the ground so that the number of pollen grains which fell could be compared with the number caught on slides exposed at the height of the plants.

During 30 minute periods on May 26 when light breezes swept across the field a large number of pollen grains were caught on the aerial slides. Only a small number was caught during periods of calm. Considerable variation is shown in the number of pollen grains caught during different 30 minute intervals of this relatively calm day (Fig. 6). Almost four times as many pollen grains were caught on the ground exposures as on the aerial exposures.

A wind velocity of 7 m.p.h. on May 29 resulted in the pollen being dispersed immediately upon dehiscence of the anthers. The data indicate an increase in the amount of pollen shed each 30 minute period until the peak of production was reached at 6:30 A.M. A rapid decrease in amount occurred until shedding ceased around 11:00 A.M. (Fig. 6). Considerably less pollen was deposited on the slides exposed on the ground on May 29 than on May 26. Thus more pollen is dispersed into the air on windy days, which probably is the reason hay fever susceptibles often are more uncomfortable on such days.

Other species: Related species within a genus were characterized by similar pollen shedding habits (Table 1). Blooming and pollen shedding occurred at approximately the same time of the day for five species of wheatgrasses, as well as for the closely related Russian wild rye. Species within the genera, *Ambrosia*, *Andropogon*, *Bouteloua*, *Festuca*, and *Sorghum* also tended to shed pollen at similar times of the day and year.

DISCUSSION

The major factors contributing to a plant's importance as a hay fever pest are length of season of bloom, abundance of pollen, intensity of pollen production and buoyance of the pollen. The small pollen grains of Mexican firebush, smooth brome grass, giant ragweed, and western ragweed, all heavy pollen producers, are very buoyant and easily dispersed by wind. The pollen of rye is considerably larger and less buoyant. Corn and sweet corn have the largest and least buoyant pollen of any grass.

A determination of the time of day of pollen shedding for the different hay fever species in a given region would aid materially in associating a patient's hay fever symptoms with certain species. Also, a measure of the amounts of pollen produced in different geographic regions by a species or by different species should throw light on the subject of why some treatments fail in one region and not in another.

SUMMARY AND CONCLUSIONS

1. Investigations were conducted to determine size of pollen and the time of day and time of year of pollen shedding of 45 species of plants when grown at Lincoln, Neb., in 1945 and Stillwater, Okla., in 1946 and 1947.

2. Mean diameters of pollen grains are reported as an aid in determining pollen size and buoyancy.

3. A determination of the time during each day when a species sheds pollen will facilitate the collection of large quantities of pollen for use in clinical medicine.

4. Petroleum jelly-coated microscope slides were so exposed as to intercept air-borne pollen. The number of pollen grains caught during 30 minute intervals per 16 sq. mm. area was used as a measure of the amount of pollen shed. This means of pollen collection gives a reliable estimate of the relative amounts of pollen produced and dispersed by species at various intervals during the day.

5. Between 90 and 99 per cent of the pollen shed was liberated by most species within a short period of three to four hours each day. The time of day of shedding varied widely for the different species and appeared to be controlled by factors inherent in each species.

6. The effects of environmental factors on pollen shedding were considered. Temperatures of less than 60° F. inhibited shedding in some species. Wind movement was important in speeding up anthesis, dehiscence, and dispersal. High relative humidity delayed pollen shedding, but did not inhibit it.

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