POLLEN PRODUCTION IN TWELVE STANDARD HARD RED WINTER WHEAT CULTIVARS

by

MIR NAJAF ALI KHAN

B. Sc., Osmania University, India, 1959
B. Sc. (Agri.), Osmania University, India, 1962

A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Agronomy

KANSAS STATE UNIVERSITY
Manhattan, Kansas

1968

Approved by:

Major Professor

TABLE OF CONTENTS

INTRODUCTION

The possibility of hybrid wheat was stimulated when Kihara (1951) developed cytoplasmic male sterility in wheat through reciprocal crosses between Aegilops and Triticum. However, the first useful sterile line was produced by Wilson and Ross (1961) with Triticum timopheevi cytoplasm. The main problem in the production of the hybrid wheat is the restoration of fertility (Livers, 1964). Besides this, other problems, such as, seed set on A-lines and the ratio of the male:female lines have to be worked out. This could be aided by knowing the number of pollen grains produced in relation to the number of ovules. The ratio is wide in the case of cross fertilized species, as corn and other grasses. This is narrow in the case of wheat--being a self fertilizer. An effective way has to be sorted out for transferring the small amount of pollen to the "A" lines from the "R" lines (Heyne, 1964).

For successful cross pollination both male and female have to bloom simultaneously, the male to disperse pollen and female to receive it. Duvick (1965) (as reported by Livers) said that lodicules do not play a prominent role in the opening of the floret of wheat. But Pederson and Jorgensen (1966) and Percival (1921) concluded that the closed flowering in wheat was due to small size of the lodicules. Maheshwari and Rangaswamy (1965) said longivity of pollen is another important factor in successful cross pollination of any crop.

This thesis reports on the following problems in wheat:

1. Determination of the number of pollen grains produced in each anther and among cultivars.
2. Correlation studies between anther size and the number of pollen grains produced.
3. Lodicule size in different cultivars.

REVIEW OF LITERATURE

1. Pollen Production

Cahn (1925) found that in two wheat cultivars length of the anther had a close relationship with the number of pollen grains present. He said that pollen bearing capacity is a good index for the selection of superior strains by the plant breeders. He reported that Obenmeyer found several strains of wheat with degenerated anthers producing a scant quantity of pollen.

Rajiki and Rajiki (1966) reported from Travensin and Gorin that there was about 1000 pollen grains produced per anther of wheat. Their investigation showed that soon after the dehiscence of the anther an average of 100 to 200 pollen grains were sticking to the stigma and also deposited below the ovary.

Nigmatulin (1965) showed that scarce amount of pollen grains affected the progeny. When two pollen grains were used per flower in the fertilization of two durum varieties there was no vigor manifested in the progeny.

Gurevic (1952) showed that pollination with a large amount of pollen gave material that varied in form and mostly were semi-awned type. When few pollen grains were used the morphological characters were more constant and mostly of the awned type.

Hiemenz (1956) obtained seedlings with high vigor when moderate amount of pollen was used, compared with seedlings which were the result of use of many pollen grains at pollination time.

Maheshwari and Rangaswamy (1965) reported that several investigators obtained better germination in culture from a denser sowing of the pollen grains than with a light sowing. They reported that Tisin stated that development of the cotton boll was better when many pollen grains were used in pollination.

Rajiki and Rajiki (1966) said that the greater the number of pollen grains per ovule the higher the seed set. The average weight of the grain increased by using more pollen grains.

2. Description and Development of Pollen Grain

Wodehouse (1935) gave a detailed account of the description of grass pollen. He said that pollen grains of the Graminae are fairly uniform. The characteristic feature of grass pollen grains is the single germpore. The exine is rough and thin and protects the grain from dessication, destruction by light and other mechanical injuries. It helps in the emergence of the pollen tube and helps pollen in adapting to the environmental changes. Thus the thin, rough exine, thick internal intine, small-sized single germpore are characters of the wind pollinating species.

Percival (1921) said the development of the subepidermal layer gives rise to the sporogenous and the tapetal cells. The tapetal layer furnishes the nutrition to the developing grains. Four pollen grains are produced from each pollen mother cell. Ultimately each one of them produces its own intine and exine. The tapetum disintegrates as the anther reaches maturity.

James and Lund (1965) found that the tapetum of the pollen mother cell producing fertile pollen grains is narrow, measuring about 4-16 μ, while that producing the aborted one has a broad tapetum measuring about 28 μ.

Joppa et al. (1966) gave the major differences between the development of fertile and aborted pollen grains in wheat. Until the formation of the exine and intine there is no difference in the development of the two. The tapetal layer of the sterile pollen mother cell continues until flowering time, whereas that of fertile lines the tapetal layer disappears after the formation of the pollen grain. The most common difference is the presence of starch in the fertile pollen grain. They explain the reason for lack of starch as the poor development of the vascular tissue resulting in failure of the transport of the nutrients to the stamen.

3. Size of the Pollen Grain

Wodehouse (1935) reported that size and shape of the pollen grains are affected by the internal environment of the anther and the external climatic factors. The diameter of the wheat pollen grain given by him was 47-57 μ and that of the germpore as 6-9 μ. He described the shape as irregular, tending to be ovoid.

Blanco (1950) suggested that there was no significant influence of environment on the pollen size of corn. The pollen size is one way to analyze the inheritance of quantitative characters.

Mamelicalvino (1956) suggested that the diameter of pollen grain is directly proportional to the number of chromosomes, especially when elementary 2n species are concerned.

Forlani (1953) gave the average pollen grain size of the different wheat species as follows:

Chromosome numbers of the species	Size of pollen grain μ
2n	45.23
4n	50.44
6n	58.87

Cetl (1961) measured the pollen grain size from 2n, 4n, and 6n species and found that size increased with the number of chromosomes. The influence of different genomes was 62 percent for B, 29 percent for D, and 18 percent for G, compared with 100 percent for A.

Magoon et al. (1958) said that induction of polyploidy increased the pollen size. The amount of increase varies with the plant in consideration.

Rajiki and Rajiki (1966) gave the diameter of the fertile and the sterile pollen grains as follows:

Line	Diameter of pollen (μ) Fertile	Diameter of pollen (μ) Sterile	Line	Diameter of pollen (μ) Fertile	Diameter of pollen (μ) Sterile
A2	45.91	44.71	B2	57.55	49.56
A5	47.94	45.65	B5	51.68	47.94
A7	46.92	46.41	B7	53.99	50.83

Blanco (1950) showed that the pollen grains of inbred lines of corn had a smaller range in size but larger mean size than the pollen of cultivars and hybrids. Parents had a unimodal distribution but the F_1 had the bimodal distribution.

4. Pollen Sterility

The only work regarding the genetic cause of the pollen abortion was reported by Sears and Loegering (1961). They identified a gene Ki located on chromosome 6B of Chinese Spring wheat. This gene (Kiki, or KiKi) results in the early abortion of the pollen grains. KiKi microspores produced by KiKiki trisomics were also aborted. This gene is linked with stem rust resistance with a recombination value of 10.5 percent and with the gene for leaf rust resistance in Pawnee with a value of 10.5 percent.

Fukasawa (1963) stated that sterility in emmer wheat pollen resulted from self duplicating of the cytoplasmic particles which inhibited amino acid activation by A.T.P. and gives rise to the aberrant proteins.

Yamamoto (1956) said that an amphidiploid of rye and wheat produced 20 percent abortion in pollen in F_2 generation but it was reduced to 11.24 percent in F_3.

Kataro (1961) said that starch in pollen of einkorn wheat appears after the translocation of two nuclei, toward the germpore. He classified the pollen grains containing densely packed starch as "Starch stage" grain and those with decreased starch as "Sugar stage" grain, grain turned dark red but those containing less starch or sugar stain light red or orange red.

Magoon et al. (1958) said that the fertility of the pollen grain in Solanaceous species decreased as the number of chromosomes increased. He said there was no problem due to low fertility of pollen because 70-94 percent seed set was possible with only 20-50 percent fertile pollen grain. They concluded that pollen fertility depended on the physiological, environmental, and most important, the genetic factors.

Rajiki and Rajiki (1966) said that fertile pollen grain of wheat should stain dark when tested with iodine or aceto-carmine and should have fine granular cytoplasm. The sterile pollen grains were not oval compared to the fertile ones and they did not stain dark or have the fine granular cytoplasm.

Allard (1960) said that the male sterile condition often is recessive. Many male sterile lines have been produced in the case of barley and tomatoes. As pollen dispersal is poor in barley, seed set was low in the male sterile barley.

5. Factors Affecting Pollen Production

Pollen grains may be fertile but if they do not have the proper environment they do not germinate and are not fertile in terms of fertilization. Sambamurthy _et al_. (1956) found that the pollen grains in banana gave a positive test for fertility with iodine but they usually had a low germination percentage.

Maheshwari and Rangaswamy (1965) said that factors affecting the germination of pollen grains are: moisture, inorganic elements, and sources of energy. Some of these requirements are met from the reserves of the pollen grain but most of the time one is limiting. Excessive moisture is deleterious to pollen germination, and a common experience has been that pollination soon after rain or dew is frequently infructuous. It is known that boron is involved in the incorporation of carbohydrates in the pollen tube membrane. Boron also is essential for the synthesis of the pectins necessary for the middle lamella of newly formed cells in the pollen tube. Calcium also has been found to be necessary for the germination and the growth of the pollen tube.

Visser (1956) said that presence of optimum osmotic pressure is necessary for the germination of the pollen grain. The secretion by the other pollen grains is also necessary for the successful germination.

Lju (1960) showed that pollen grains secreted a liquid upon contact with the other pollen grains which in turn promotes the growth of the pollen tube. The damaged pollen grains also help in the pollen tube growth.

Pisarev and Zilkina (1955) said spraying with boric acid solution preceding shooting helped produce more spikelets per spike as well as an increase in seed set. This probably was due to the higher pollen germination.

6. General: Pollen Grains

Batikyan and Cholakhyan (1964) said that the secondary pollination had a definite effect on the progeny of wheat through direct fertilization of the ovary which did not get fertilized because of the primary fertilization or through the metabolic influence of the pollen grains on the growth of the pollen tubes and the sperms which were dissolved in the embryonic sac. The longer the interval between the two pollinations, the weaker the force of the male component for conferring its traits on the progeny.

Ostroverkhov (1964) said that supplementary pollination in wheat increased the seed set. Increased seed set was the result of the fertilization of the ovules not fertilized before.

Bhatnagar et al. (1964) obtained an increase in the number of spikelets per ear by transplanting wheat. Fertile florets also increased through this method.

Wheat yield was reduced by a decrease in soil temperature (Petrova, 1965). He said that roots developed slowly at a low temperature; growing season was prolonged; number of fertile florets decreased; and usually the third floret of wheat, which is fertile in most of the cases, was aborted.

With special experiments it has been shown in wheat that frost damaged the reproductive organs. The resistance of the crop decreased as the flowers developed. The most sensitive part of the flower is the androecium. Frost affects the metabolism of the plant causing an accumulation of fat-like substances in the androecium and hampers the production of the pollen (Petrova and Drozdov, 1964).

7. Lodicule Size

Duvick (1965) said that opening of the flowers did not depend on the lodicule size. But Pederson and Jorgenson (1966) showed that lodicule size was closely linked to the opening of the flowers and the closed flowering was correlated with small sized lodicules.

Percival (1921) said that the lodicules become turgid assuming a convex shape at the time of anthesis. They push apart during anthesis and help in opening of the florets. When all the anthers are carried into the air, the florets close and lodicules become flaccid. The anthers were pushed out in compact ears in the florets of the lower spikelets and in the small florets of the laxial ears.

MATERIAL AND METHODS

Twelve standard cultivars (Table 1) of wheat were grown in a complete randomized block design on the Agronomy Farm, Kansas State University, Manhattan. Fifteen spikes were selected at random for each cultivar and preserved in 70 percent alcohol. The collection was completed in three days, as all the varieties bloomed within this time. The time for collecting these heads was important. Spikes containing fully developed anthers but not yet dehisced were selected. The date of collection was from 18th May to 20th May, 1966. The blooming was later than average in 1966. A freeze on 13th May affected the wheat plant to a considerable extent.

Wheat spikes, on an average, consisted of 12-15 spikelets. Each spikelet consisted of three florets, two primaries and one secondary. Generally the secondary florets were sterile, but some set seed. However, the anthers were

Table 1. Cultivars included in the study of pollen number and size.

Variety	CI No.	Origin or pedigree
Turkey	1558	An Introduction
Gage	13532	Ponca 3/ Mediterranean / Hope 2/ Pawnee
Bison	12518	Chiefkan 2/ Oro / Tenmarq
Triumph	12132	Kanred / Blackhull 2/ Florence 3/ Danne Beardless / Blackhull
Ottawa	12804	Mediterranean / Hope 2/ Pawnee 3/ Oro / Ill. No. 1 / Comanche
Kaw	12871	Early Blackhull / Tenmarq 3/ Oro 2/ Mediterranean / Hope
Wichita	11952	Early Blackhull / Tenmarq
Ponca	12128	Kawvale / Tenmarq 2/ Kawvale / Marquillo
Comanche	11673	Oro / Tenmarq
Scout	13546	Nebred 2/ Hope / Turkey 3/ Cheyenne / Ponca
Parker	13285	Quivira 3/ Kanred / Hard Federation 2/ Prelude / Kanred 4/ Kawvale / Marquillo 2/ Kawvale / Tenmarq
Triumph 64	13679	Danne Beardless / Blackhull 3/ Kanred-Blackhull / Florence 2/ Kanred-Blackhull / Triumph

generally smaller and contained less pollen than the primary florets and the seed was two-thirds the size of the kernels found in the primary florets.

Each floret consisted of the lemma, palea, three stamens, and one pistil consisting of the feathery stigma and the ovary. Allard (1960) said that flowers in wheat are structurally similar to those of rye, but in wheat the anthers burst before extrusion so that the empty anthers hanging out are only a mockery. The chasmogamous behavior of wheat flowers does, however, permit a low incidence of out crossing.

In the present study the anthers were sampled from the middle spikelets. Five to six spikelets in a spike constitute the middle portion of the spike. Generally the top spikelet is sterile and in most of the cases the bottom one too.

1. Pollen Counts

A single anther was cut longitudinally into halves on a microscopic slide. The pollen from one half was dispersed into water and then distributed as uniformly as possible. The pollen grains present in the half anther were counted under the dissecting microscope through the divided eye piece. The divided eye piece helped to divide the field in different squares. The pollen in each square was counted and then multiplied by two to get the total amount present in the single anther. The time required to count the pollen in one-half of the anther was approximately five to seven minutes. On the average, twelve anthers were counted per hour and about eighty could be counted in eight hours.

Cahn (1925) counted pollen grains by dispersing pollen on a glycerine smeared slide which was marked and divided. This was not applicable in the present study because the spikes were preserved in 70 percent alcohol and the anthers were wet and the pollen did not come out when shaken. Counts also were tried by dispersing the pollen into other media. This gave error in counts because the pollen was not uniformly distributed throughout. Pollen was heavy and settled to the bottom of the medium.

2. Anther Size

A 2x eye piece and 5x objective were used for measuring the anther length and width. The higher magnification of either one would make the anther to go out of the scale lengthwise. A standard scale was used for measurement of the length and width. The scale was divided into 150 divisions. All these standard measurements were converted later into millimeters.

A micrometer scale with .01 mm divisions was set on the microscope stage. The standard scale was fixed in the eye piece of the microscope. The two scales were parallaxed at one position. At one position it was found that five divisions of the standard scale equaled fifteen divisions of the micrometer. Therefore, one division of the standard scale was equal to three divisions of the micrometer. Since each division of the micrometer was equal to .01 mm, therefore each division of the standard scale was equal to .03 mm. All the measurements of the standard scale were multiplied by .03 to convert to the millimeter scale.

Anther size was calculated by multiplying the length with width, assuming the anther to be rectangular in shape.

The lodicule length and width were measured on the same standard scale. Only the effective length of the lodicule was measured and the width in the middle was measured. The size of the lodicule was calculated by multiplying the length with the width and a constant factor of $\pi/4$; the shape of the lodicule was assumed to be elliptical (Selly and Brain, 1965).

RESULTS AND DISCUSSION

Six spikes were sampled from all cultivars, except for Triumph 64, where only five were sampled. The remaining spikes collected were either too early or they had already shed their pollen.

The mean of the pollen number for the twelve cultivars is given in Table 2.

Table 2. Average pollen number per anther of twelve winter wheat cultivars. Manhattan, Kansas, 1966.

Cultivar	Mean
Ponca	1629 ± 318.2
Gage	1595 ± 195.5
Kaw	1489 ± 182.5
Scout	1465 ± 148.6
Comanche	1445 ± 204.8
Wichita	1429 ± 229.9
Ottawa	1378 ± 159.8
Turkey	1332 ± 137.9
Parker	1324 ± 100.6
Bison	1311 ± 134.0
Triumph	1294 ± 165.8
Triumph 64	1282 ± 191.4

LSD .05 = 217.4

The number of pollen grains among the cultivars was statistically significant. Ponca had the highest number (1629) of pollen grains and Triumph 64 the lowest (1282). Triumph 64, Triumph and Bison were significantly different from Ponca and Gage (LSD .05 = 230).

Rajiki and Rajiki (1966) reported from Taravensin and Gorin that an anther of wheat contained an average of about 1000 pollen grains. The figures obtained in the present study are from anthers sampled from the middle spikelets. When all three positioned spikelets were sampled in the cultivar Ottawa, the number was about 1250. In general, the topmost and the last spikelet in the spike did not have the reproductive organs.

Rajiki and Rajiki (1966) reported that the seed set in the middle spikelets was higher than in the top and the bottom spikelets because of greater number of pollen grains. From the figures obtained in this study it could be calculated that approximately 4000 pollen grains are available per ovule in the middle spikelets and on an average about 3000 pollen grains per ovule are available in the top and the bottom spikelets. This amount would still be reduced during hybrid wheat program because, according to Taravensin and Gorin, soon after the anthesis of the anther 100-200 pollen grains are found attached to the stigma of the pistil. Many of the pollen grains also are found lying below the ovary. Such losses also were observed in this study. If a statement made by Allard (1960), i.e., empty anthers hanging outside are a mockery, is considered correct, then none of the pollen present in the wheat would be available for cross pollination. But some amount could always be found available in the study made at Kansas State University in 1966.

On the average, a single spike of wheat was found to contain about twelve spikelets. The number of spikelets per spike in the cultivars studied was non-significant. Considering only primary florets, each middle spikelet would produce about 8000 pollen grains and the top and the bottom spikelet would produce about 6000 pollen grains. It has been found under this study that six spikelets in the middle could, on the average, produce about the same number of pollen grains. Therefore, about 48,000 pollen grains could be available from the middle spikelets only. The remaining six spikelets from the top and the bottom could produce on the average 36,000 pollen grains. Therefore, a single main culm of a wheat plant could produce about 84,000 pollen grains, or about 3400 pollen grains per ovule. This is meager when compared with the 50,000 pollen grains produced per silk in corn. Even if the pollen grains present in a single anther are available for pollinating a single spike, i.e., 1200 pollen grains, and considering that only 20 percent of the pollen is available for pollination during cross pollination, then about 15-20 main shoots of the wheat could be fertilized with a single shoot of the male. If the pollen amount produced by the tillers was known, then one could calculate roughly the male:female ratio in wheat. This has been tried by the DeKalb research station with a ratio of 1:2 (Holland, 1966).

During the study only the primary florets were sampled. In a majority of the cases the secondary florets were found to contain pollen grains. The anther size and the number of pollen grains were less than those in the primary florets.

The figures given in this study represent both fertile and the aborted pollen grains. Apparently, Ponca contained the highest percentage of pollen abortion--about 4 to 5 percent. Other cultivars had only one or two percent pollen abortion.

One of the primary florets develops earlier than the other, so if one floret was found to contain dehisced anthers it was no indication that the other one was also dehisced.

Petrova and Drozdov (1964) said that the anthers are the most sensitive parts of the stamens affected by frost. The cultivars under study were damaged by frost on 13th May 1966. The anthers of the cultivars as Cage, Turkey, Bison, and Comanche were found to be affected the most. It was not possible to count the pollen grains in such anthers as they were full of tissue material. These damaged anthers did not dehisce. It was interesting to note that frost had not attacked the anthers of the same spikelet equally; this again shows that rates of development of the florets are different. The most frost damage was in Turkey and Bison cultivars.

The size of the pollen grain varied. The pollen grain size as well as the anther size of the primary florets from the top and the bottom spikelets was smaller but the number of pollen grains was similar to that produced in the middle spikelets.

The anther size in Parker was small compared to the other cultivars. It contained pollen numbers equal to Bison whose anther size was large. This showed the effect pollen size had on the number of pollen grains. Pollen of Kaw, Bison, and Ottawa was large but that of Ponca was small. That is why, though the anther size of Ponca was smaller than Gage, Bison, and Comanche, it contained the greatest number of pollen grains of these cultivars.

It was noticed a number of times during this study that one could guess the size of the pollen by seeing the spikelet size. Most of the time the two factors are directly proportional. Sometimes when the pollen was not matured this would not hold true. The mature pollen grain could be identified by KIK solution. Mature pollen would turn violet and the immature pollen would be brown. This made it possible to fix a particular stage for the sampling of pollen grains. In the field the samples should be collected when the head or spike of wheat comes out of the boot leaf, because at this time pollen grains are fully formed and mature.

Ponca gave the highest number of pollen grains, yet its standard deviation was the highest among all cultivars. This might be due to the variability in pollen size which affected the pollen number.

Parker had the lowest deviation, thus showing the uniformity of the development of the anther. Parker was one of the early cultivars.

The amount of pollen grains differed in the individual anther within the same floret. This might be due to the difference in the development of the pollen grains in the anther which was greatly affected by the internal environment of the anther (Wodehouse). This was prominent in the case of Gage where it was observed that one anther had produced 1500, whereas another in the same floret produced as many as 2000 pollen grains. Ottawa was peculiar in this regard as it produced different amounts of pollen grains in the anther of the same floret.

Cultivars having a similar parentage were found to be statistically non-significant with regard to pollen production. For example, Scout and Gage have Ponca in their parentage. Similarly, Bison and Comanche have Oro/Tenmarq in their parentage. Kaw and Wichita have Early Blackhull/Tenmarq in their parentage, and also were statistically not different.

Anther Length

The analysis of variance was calculated with the standard scale measurements. The means of anther lengths are given in Table 3. They were converted from the standard scale by multiplying them with a factor of .03. The anther length was statistically significant both at 5 and 1 percent levels of significance. Comanche anthers were the longest and Parker the shortest. Ponca had the largest number of pollen grains which was significantly different from Ottawa. However, its anther length was not significantly different from Ottawa. Four groups could be made among the cultivars with regard to this character. Parker was significantly

Table 3. Anther lengths in millimeters of the twelve winter wheat cultivars. Manhattan, Kansas, 1966.

Cultivar	Mean
Comanche	3.708 ± .1500
Bison	3.666 ± .1631
Gage	3.633 ± .2733
Ponca	3.606 ± .2865
Turkey	3.588 ± .2650
Kaw	3.585 ± .1319
Scout	3.480 ± .2145
Ottawa	3.363 ± .2010
Wichita	3.210 ± .2793
Triumph	3.210 ± .1242
Triumph 64*	3.060 ± .3570
Parker	2.825 ± .0732

LSD .05 = .2513

*Triumph 64 is nonsignificant with Wichita, Triumph, and Parker. (LSD = .2639)

smaller than all others. The cultivars from Comanche to Scout form one group, Ponca to Ottawa one, and Ottawa, Wichita, and Triumph form one group.

Anther length was found to be more variable within a cultivar than pollen number. For example, one spike of Turkey had an average of 3.29 mm for anther length, while another had 3.94. The standard deviations of Gage, Ponca, and Wichita were high showing the variability within a cultivar. Parker had the smallest standard deviation indicating uniformity within the cultivar.

The anthers of Parker were small and could be easily identified by this character. The spikes also were small compared to the other cultivars. Comanche had anthers of uniform length. The high standard deviation of Triumph 64 was due to the fact that the spikes were at different stages of development though they were collected on the same date.

Cahn (1925) said there was a strong correlation between anther length and the pollen number. This also was observed in the nine varieties out of the total under study. However, Cahn did not take into consideration the pollen size. Pollen size strongly affected the correlation when the individual spike correlations were calculated.

Correlation between anther length and number of pollen grains is given in Table 4. Overall correlations for all varieties were low (.3899) and non-significant. This could be due to the cultivar effect and also due to the size of the pollen number.

However, one could infer that the longer the anther length the greater will be the pollen number. The correlations discussed above were calculated by taking the means of the pollen numbers and the anther length of the individual spikes.

Table 4. Correlation coefficients between anther length and pollen number of twelve cultivars grown at Manhattan, 1966.

Cultivar	Anther length (mm)	No. pollen per anther	Correlation (Mean of the pollen no. and anther length of the six individual spikes)
Turkey	3.57	1332	.930**
Gage	3.63	1595	.846*
Bison	3.66	1294	.939**
Triumph	3.21	1311	.900*
Ottawa	3.36	1378	.898*
Kaw	3.60	1489	.353
Wichita	3.36	1429	.665
Ponca	3.60	1629	.903*
Comanche	3.69	1445	.672
Scout	3.48	1465	.965**
Parker	2.94	1324	.865*
Triumph 64	3.06	1282	.908*

If we examine this table, we find that Kaw and Ponca have the same anther length but there was a difference in the pollen number that affected the correlation.

Similarly though Bison and Parker are statistically non-significant in pollen number; their anther lengths are different which affected the correlations. Bison and Gage are statistically non-significant in anther length but were significantly different in pollen number.

Table 5. Mean of the anther width, in mm, of the twelve cultivars grown at Manhattan, Kansas, 1966.

Cultivar	Width
Wichita	.726 ± .063
Comanche	.681 ± .087
Gage	.669 ± .139
Kaw	.669 ± .028
Ponca	.663 ± .055
Scout	.663 ± .131
Bison	.648 ± .031
Ottawa	.636 ± .047
Turkey	.632 ± .149
Parker	.624 ± .034
Triumph 64	.615 ± .044
Triumph	.603 ± .054

LSD .05 = .058

The anther width of the twelve cultivars is given in Table 5. The width of the anther was significantly different at 5 percent with an LSD of .058. There were three groups. Wichita had the highest value (.726) and Triumph the lowest (.603). This character was uniform. In fact there were very minor differences among the cultivars. One peculiarity with this character was that there was not much difference in the positions of the spikelets. Anthers in the upper and lower spikelets most of the time had widths equal to that of the middle spikelets. Due to the uniformity, the deviations from the means are not high, except in a few cases. Because of this, one could infer that it is not necessary that if the length is long, then the anther should be wider too. Correlations calculated between length and width, taking the overall means of length and width, gave non-significant results.

Correlation values between widths and pollen number were low (Table 6) with Turkey, Scout, Parker, and Comanche, only two values being significant. Cultivars as Ottawa, Scout, Parker, and Gage, which showed high correlations between length and pollen number, gave low values, thus showing the marked effect of length on the amount of pollen grains. Kaw showed a high correlation value between anther width and pollen number indicating the effect of width. Wichita was similar. Comanche was the only cultivar which did not show either significant correlations between length and pollen or width and pollen. The overall correlation value was also non-significant.

Table 6. Correlation between anther width and pollen number of the twelve cultivars. Manhattan, Kansas, 1966.

Cultivar	Correlation (Mean of the anther width and pollen no. of six heads)
Turkey	-.048
Gage	.400
Bison	.793
Triumph	.798
Ottawa	.406
Kaw	.709
Wichita	.855*
Ponca	.711
Comanche	.211
Scout	.137
Parker	.462
Triumph 64	.860*

Regression Analyses between Anther Length and Pollen Number

Statistically, regression is defined as the unit increase or decrease in X, there will be corresponding increase or decrease in Y. Therefore, if originally the anther measured 3.00 mm in length, and there was an increase to 4.00 mm and the original anther had 1300 pollen grains, the longer (4.00 mm) anther would have 1750 pollen grains. The regression values between anther length and pollen number are given in Table 7.

Table 7. Regression analysis between anther length and pollen number of twelve cultivars. Manhattan, Kansas, 1966.

Cultivar	Regression	Regression x .01
Turkey	554.76**	55.476
Gage	613.27*	61.327
Bison	764.04**	76.404
Triumph	1195.38**	119.538
Ottawa	658.99**	65.899
Kaw	452.00	45.200
Wichita	547.71	54.771
Ponca	903.71**	90.371
Comanche	924.00	92.400
Scout	667.90**	66.790
Parker	1206.00**	120.600
Triumph 64	490.60*	49.060

These values show that increased length of the anther where pollen size was small would increase the number of pollen grains, e.g., Triumph, Ponca, and Parker.

<u>Regression Analysis between Anther Width and Pollen Number</u>

As the increase in anther width was in terms of .01 units, the calculation on the basis of increase in a unit basis gave large numbers. The regression values are presented in Table 8.

Table 8. Regression values between anther width and pollen number of twelve cultivars of winter wheat. Manhattan, Kansas, 1966.

Cultivar	Regression value x(.01)
Turkey	-.260
Gage	5.90
Bison	42.67
Triumph	21.84
Ottawa	13.94
Kaw	47.19
Wichita	30.60
Ponca	43.60
Comanche	7.03
Scout	1.55
Parker	13.67
Triumph 64	33.33

Increased width resulted in only small increases in the amount of pollen.

Ponca, Kaw, Triumph 64, Bison, and Wichita had higher regression values indicating that pollen number could be increased by increasing the anther width in these cultivars.

The mean anther size, determined by length x width, of the twelve cultivars is given in Table 9. Comanche had the largest size index and Parker the smallest. Comanche also had the longest anther length and Parker the shortest.

Table 9. Means of the anther size of the twelve standard cultivars of winter wheat. Manhattan, Kansas.

Cultivar	Mean
Comanche	2.547 ± .307
Wichita	2.455 ± .429
Gage	2.433 ± .116
Ponca	2.431 ± .387
Kaw	2.358 ± .151
Bison	2.358 ± .171
Scout	2.312 ± .285
Turkey	2.267 ± .280
Ottawa	2.165 ± .293
Triumph	1.934 ± .223
Triumph 64	1.896 ± .233
Parker	1.844 ± .130

LSD .05 = .3330

Wichita had the widest anther width, thus indicating that width did have some effect on anther size index.

Both Triumph and Triumph 64 are similar to Parker for anther size index; both their lengths being slightly larger than Parker.

There was one large group of eight cultivars which were non-significant for anther size index.

Correlations and regression values between anther size and pollen number are given in Table 10.

Table 10. Correlation and regression values between anther size and pollen grains of twelve winter wheat cultivars. Manhattan, Kansas, 1966.

Cultivar	Correlation	Regression x .1
Turkey	.236	11.37
Gage	.285	15.95
Bison	.979**	74.60**
Triumph	.829*	60.78*
Ottawa	.771	42.15
Kaw	.748	92.67
Wichita	.808*	43.64*
Ponca	.973**	80.08**
Comanche	.530	35.36
Scout	.717	37.43
Parker	.828*	65.02*
Triumph 64	.802*	29.12

In most cases, the significant correlations between anther size index and pollen number were due to the effect of anther length on the pollen number except for Wichita and Kaw, where correlation between length and pollen number was non-significant. However, as width had the same effect, correlation between anther size index and pollen number were significant in Wichita and high in Kaw.

Comanche had low correlation values for length, width, and anther size index with pollen number suggesting that pollen size may have a pivotal role.

Lodicule size was determined for ten cultivars and the averages given in Table 11. The results are significant both at 5 percent and 1 percent. The LSD at .05 was .078. Duvick (1965) said that lodicule size did not play a significant role in the opening of the flowers.

Table 11. Lodicule size of the ten winter wheat cultivars. Manhattan, Kansas, 1966. (LxWx π/4)

Cultivar	Lodicule size
Scout	.763
Comanche	.669
Ponca	.610
Kaw	.582
Ottawa	.579
Gage	.572
Triumph 64	.560
Bison	.544
Parker	.542
Triumph	.516

LSD .05 = .078

The relationship between closed flowering and small sized lodicules mentioned by Percival (1921) and Jorgenson and Pederson (1966) might be in the ordinary self fertile wheat where the lodicules do help in the opening of flowers.

Hayes *et al*. (1955) mentioned that glumes of self unfruitful florets are held open by the lodicules and rapid application of the desired foreign pollen by dusting on the exposed stigma was possible.

There were no great differences among the cultivars for lodicule size. Scout had the highest value and Triumph the lowest.

Opening of the florets by the lodicule size would not help in a self fertilized plant like wheat, because by the time the flower was mature and open, and anthers were dehised, pollination already had taken place.

In artificial fertilization this could be helpful, be cause the florets remained open about two days if they were not pollinated.

It was noticed that after closing of florets due to contraction of lodicules, they opened again if they were not fertilized. The exact mechanism of the opening of the flower for the second time is not fully understood.

SUMMARY

Even though Kihara demonstrated cytoplasmic male sterility in 1951, there are many problems to be solved before successful hybrid wheat can be commercially produced. One factor is the ratio of the male and female lines. For this, among many other factors, the amount of pollen produced by each anther is an important factor.

The amount of pollen produced per anther was studied in twelve cultivars grown at the Agronomy farm in 1966 in a complete randomized design. The other characters studied were anther size and lodicule size.

Correlations were calculated between anther length and pollen number, width and pollen number, and anther size and pollen number for each cultivar. Many correlations were significant between anther length and pollen number and anther size and pollen number. There were only two correlations significant between anther width and pollen number.

All the characters measured were statistically significant among the cultivars studied.

LITERATURE CITED

1. Allard, R. W. (1960)
Principles of Plant Breeding. Wiley and Sons Co. p. 33.

2. Batikyan, G. G. and Cholakhyan, D. P. (1964)
The influence of secondary pollination on the inheritance of traits in corn. Biol. Abs. 45:3910.

3. Bhatnagar, M. P., Bhargava, P. D., and Gandhi, S. M. (1964)
Effect of transplanting on wheat. Biol. Abs. 45:7687.

4. Blanco, J. L. (1950)
Pollen size and quantitative inheritance in maize. Genetics 35:654-55.

5. Briggle, L. W. and Reitz, L. P. (1963)
Classification of Triticum and wheat varieties grown in U. S. Tech. Bul. Agr. Res., Serial No. 1278.

6. Cahn, Erich (1925)
A study of fertility in certain varieties of common wheat with respect to anther length and amount of pollen present in parents and offspring. J. Am. Soc. Agron. 17:591-95.

7. Cetl, I. (1961)
The size of the pollen grain of the genus Triticum. Pl. Br. Abs. 31:600-01.

8. Duvick, D. N. (1966)
Influence of Morphology and Sterility on Breeding Methodology. Plant Breeding. A symposium held at Iowa State University. Edited by Kenneth J. Frey. Iowa State University Press: Ames, Iowa. pp. 85-138.

9. Forlani, R. (1953)
Observations on some artificial allopolyploids with particular reference to the size of the pollen grain. Pl. Br. Abs. 23:558.

10. Fukasawa, A. H. (1963)
Biochemical mechanism of pollen abortion and other alterations in the cytoplasmic male sterile wheat. Pl. Br. Abs. 33:563.

11. Gurevic, L. I. (1952)
The influence of pollination methods upon the process of form development, in sexual hybridization of wheats. Pl. Br. Abs. 22:376.

12. Hayes, H. K., Immer, F. R., and Smith, D. C. (1955)
Methods of Plant Breeding. McGraw-Hill Co., Inc. p. 85.

13. Heyne, E. G. (1964)
Progress with hybrid wheat. 47th Report, Kansas State Board of Agriculture. pp. 67-70.

14. Hiemenz, G. (1956)
Investigation in gene component and successive fertilization in S. viriabilis and their effect on the next generation. Pl. Br. Abs. 26:13.

15. Holland, R. F. (1966)
Hybrid wheat when. Crops and Soils 18:7.

16. James, N. I. and Lund, S. (1965)
Induction of male sterility in barley with K. gibberellate and plant growth regulators. Agron. J. 56:269-72.

17. Joppa, L. R., McNeal, F. H., and Welsh, J. R. (1966)
Pollen and anther development in cytoplasmic male sterile wheat (T. aestivum L.). Crop Sci. 6:296-97.

18. Kihara, H. (1951)
Substitution of nucleus and its effects on genome manifestations. Cytologica 16:177-193.

19. Kataro, Watanabe (1961)
Studies on the germination of grass pollen. II. Germination capacity of pollen in relation to maturity of pollen and stigma. Bot. Magazine 74:131-135.

20. Lju, D. C. [Liu, T. T.] (1960)
Microscopical observations on mutual interaction between pollen grains. Pl. Br. Abs. 30:502.

21. Livers, R. W. (1964)
Fertility restoration and its inheritance in cytoplasmic male sterile wheat. Science 144:420.

22. Magoon, M. L., Cooper, D. C., and Houges, R. W. (1958)
Induced polyploidy of Solanums and their crossability with Solanum tuberosum. Bot. Gazette 119:220-31.

23. Maheshwari, P. and Rangaswamy, N. S. (1965)
Embryology in relation to physiology and genetics. Advances in Botanical Research 7:219-310.

24. Mamelicalvino, E. G. (1956)
The correlation between chromosome number and the dimensions of the microspore in botanical taxonomy. The genetic spectrum of pollen grain. Pl. Br. Abs. 26:150.

25. Nigmatulin, F. G. (1965)
The relation between amount of pollen used in fertilization and inherited characteristics and properties in hybrids of soft and hard wheats. Biol. Abs. 46: 4006.

26. Ostroverkhov, V. O. (1964)
The effect of pollination methods on sexual process and hybrid formation in wheat. Biol. Abs. 45:6030.

27. Pederson, P. N. and Jorgenson, J. (1966)
Inheritance of size of lodicule and closed flowering in barley. Pl. Br. Abs. 36:500.

28. Percival, J. (1921)
The wheat plant. A monograph. E. Dutton Co., New York. pp. 116-119.

29. Pisarev, V. E. and Zilkina, M. D. (1955)
Using boron to stimulate the growth of hybrids of low fertility. Pl. Br. Abs. 25:165.

30. Petrova, L. R. and Drozdov, S. N. (1964)
Effects of the night frost on the formation of reproductive organs of the spring wheat. Biol. Abs. 45: 5387.

31. Petrova, L. R. (1965)
The specific characteristics of formation of summer wheat ears when the roots are cooled. Biol. Abs. 46: 4739.

32. Rajiki, E. and Rajiki, S. (1966)
Research work of hybrid wheat at Martonvassar. Acta Agronomica Academiae Scientriarum Hungaricae 15(1-2): 199-214.

33. Sambamurthy, K., Khan, K., Fazlullah, Bettan E., and Gowdar, R. (1956)
A study of pollen of South-Indian banana varieties. Pl. Br. Abs. 26:21.

34. Sears, E. R. and Loegering, W. Q. (1961)
A pollen killing gene in wheat. Genetics 46:897.

35. Selly, Samuel N. and Brain, Grinling (1965)
Standard Mathematical Tables. The Chemical Rubber Co., p. 495.

36. Visser, T. (1956)
Germination and storage of pollen. Pl. Br. Abs. 26: 155.

37. Wilson, J. A. and Ross, W. M. (1961)
Cross breeding in wheat, Triticum aestivum. I. Friquency of the pollen restoring character in hybrid wheats having Aegilops ovata cytoplasm. Crop. Sci. 1: 191-193.

38. Wodehouse, R. P. (1935)
Pollen Grains. McGraw Hill Book Co. pp. 303-319.

39. Yamamoto, K. (1956)
Duration of ovular potency and the pollen of rye wheat amphidiploids and the parents. Pl. Br. Abs. 26:396.

ACKNOWLEDGEMENT

The author wishes to express his deep appreciation to Dr. Elmer G. Heyne, Major Professor, for his advice and counsel during the preparation of the thesis. His thanks also go to Dr. G. H. Liang for allowing the use of his laboratory for carrying out the experiment, and to Dr. Olson and Dr. Goss for reviewing the manuscript and offering helpful suggestions. The author also wishes to thank the Kansas Crop Improvement Association for the graduate research assistantship during the later part of this study.

POLLEN PRODUCTION IN TWELVE STANDARD
HARD RED WINTER WHEAT CULTIVARS

by

MIR NAJAF ALI KHAN

B. Sc., Osmania University, India, 1959
B. Sc. (Agri.), Osmania University, India, 1962

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Agronomy

KANSAS STATE UNIVERSITY
Manhattan, Kansas

1968

The knowledge of amount of pollen grain produced within a single anther and hence in a single spikelet of wheat is an important aspect of the hybrid wheat development program. With the help of such information, one may assess the male-female ratio. A study was conducted in 1966 with twelve standard cultivars of wheat grown at the Agronomy farm, Kansas State University. A random sample of 15 spikes was collected in each cultivar and preserved in 70 percent alcohol.

The pollen number in each half of the anther was counted, through a divided eye piece under a binocular; the number was multiplied by two to get the total number of pollen grains in a single anther. The anther length and width were measured on a standard scale, and later the readings were converted into millimeters by multiplying with a factor of .03. The anther size was calculated by multiplying length and width. Similarly lodicule size was obtained by measuring the effective length multiplied by width and a factor of $\pi/4$.

Statistically these floral characters were significant at the 5 percent level of probability. Correlations were calculated between anther length, width, and size, and pollen number for all the twelve cultivars. Nine of the 12 correlations were significant between anther length and pollen number. Two correlations were significant between anther width and pollen number and six were significant between anther size and pollen number. Overall correlations were calculated for anther length and width. They were non-significant.