INFLUENCE OF HUMIDITY ON FLORAL INFECTION OF WHEAT AND BARLEY BY LOOSE SMUT[1]

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INTRODUCTION

In a study of the relative resistance of some wheat varieties to loose smut (*Ustilago tritici* (Pers.) Rostr.), previously reported by the writer (*12*),[3] the three club wheats Hybrid 128 (C. I.[4] 4229), Jenkin (C. I. 5177), and Little Club (C. I. 4066) proved strikingly susceptible when grown from seed from flowers that had been artificially inoculated under humid conditions. In view of the fact that loose smut occurs but rarely in the Pacific Coast States (fig. 1),

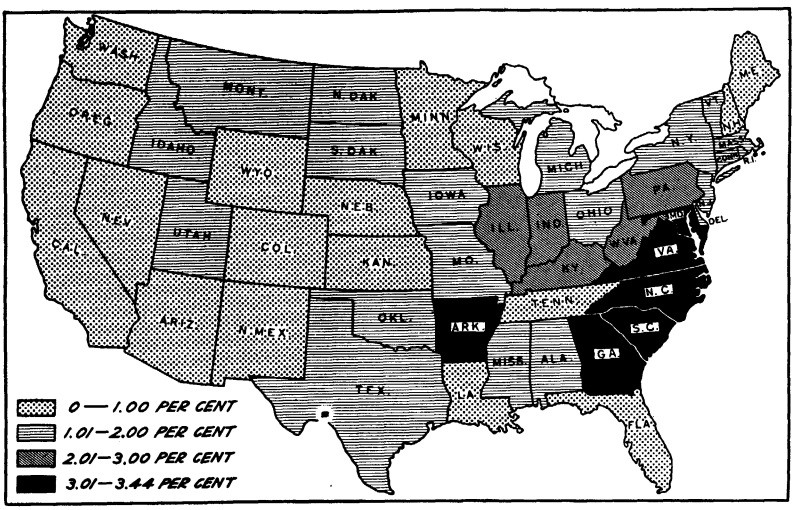

FIGURE 1.—Estimated average annual reduction of wheat production (per cent) due to loose smut in each State for the period 1917–1927. The average reduction (per cent) for each State has been placed in one of four groups. Data compiled from estimates of the Plant Disease Bulletin (footnote 6)

where these wheats are grown, it seemed probable that the low atmospheric humidity, which generally prevails there when the wheats are in bloom (fig. 2), might play an important rôle in preventing infection. The probability of this relation was strengthened by the writer's observations during the summers of 1928 and 1929 in the State of Idaho on the loose-smut content in wheats grown on

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[3] Reference is made by number (italic) to Literature Cited, p. 515.

[4] C. I. indicates accession number of the Division of Cereal Crops and Diseases.

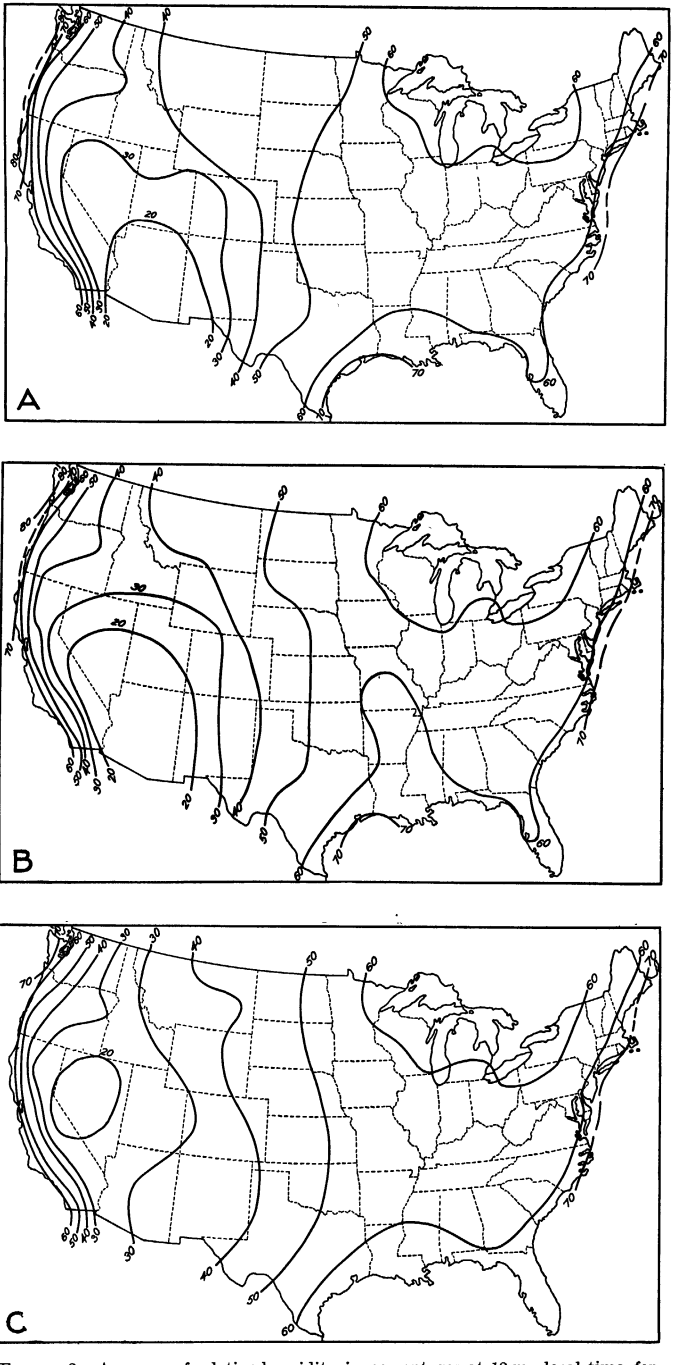

FIGURE 2.—Averages of relative humidity in percentages at 12 m., local time, for May (A), June (B), and July (C), for the 10-year period 1918–1927. Data from Weather Bureau

irrigated and on dry land. Loose smut was found prevalent and causing considerable damage in the wheats grown under irrigation but was seldom observed in the wheats grown on dry land. In this latter group the Turkey variety predominates. Three different selections of this wheat proved highly susceptible to loose smut in tests conducted by the writer (*12*).

In a study of the occurrence of *Ustilago tritici* in the United States, it was noted, furthermore, that this smut is more prevalent in the eastern than in the western half of the country.[5,6] (Fig. 1.) In this connection it is important to note that at the different periods when the wheats are in bloom [7] in the different sections in the United States the relative humidity, in general, also is higher in the eastern than in the western half of the country. (Fig. 2.)

In the United States, measures for controlling loose smut of wheat are rarely applied. Control through seed treatment is seldom employed, largely because the causal fungus, hibernating inside the seed, is not amenable to control by the easily applied surface disinfectants such as copper carbonate and formaldehyde. Treatments that thoroughly soak the seed, like the modified hot-water method, are necessary to kill the intraseminal mycelium. However, treatments of this kind are difficult to apply under the usual farm conditions. Furthermore, the modified hot-water treatment may cause severe injury to germination, thus reducing the yield (*11*). Recent studies by the writer (*12*) have shown that at least in the case of wheats grown in the Eastern States only a few commercial varieties are resistant. As a result of this situation the losses from loose smut in wheat continue unchecked.

It seemed desirable, therefore, to determine whether or not low relative humidity during the period of blooming might be the factor involved in the natural control of *Ustilago tritici*. It was felt that if a positive relation were found it might lead to a consideration of the practicability of control through the use of seed from areas with appropriately low ranges of relative humidity during the blooming period of the wheats.

The loose-smut fungus of barley (*Ustilago nuda* (Jens.) Kell. and Sw.) was also included in some of the studies herein reported. In general, the situation regarding the geographical occurrence of *U. nuda* and the hot-water treatment of the seed recommended for its control is similar to that of the loose-smut fungus of wheat.

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[5] In Idaho and Utah a considerable portion of the wheat is grown under the humid conditions accompanying irrigation, thus masking the effects of the normally low humidity on the control of wheat loose smut.

[6] UNITED STATES DEPARTMENT OF AGRICULTURE. BUREAU OF PLANT INDUSTRY. The following mimeographed material from Plant Disease Bulletin (or Reporter):

ESTIMATE OF CROP LOSSES DUE TO PLANT DISEASES. 1917. Plant Disease Bul. 2 (1), 18 p. 1918.
CROP LOSSES FROM PLANT DISEASES. 1918. Sup. 6, p. 186–213. 1919.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1919. Sup. 12, p. 307–332. 1920.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1920 Sup. 18, p. 317–338. 1921.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1921. Sup. 24, p. 489–510. 1922.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1922. Plant Disease Rptr., Sup. 30, p. 462–490. 1923.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1923. Sup. 36, p. 318–348. 1924.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1924. Sup. 43, p. 381–410. 1925.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1925. Sup. 49, p. 382–412. 1926.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1926. Sup. 56, p. 394–423. 1927.
CROP LOSSES FROM PLANT DISEASES IN THE UNITED STATES IN 1927. Sup. 64, p. 370–399. 1928.

[7] The approximate dates of blooming may be obtained from the harvest dates given (*2, 3*) by subtracting one month from the harvest dates.

LIFE HISTORY OF THE CAUSAL ORGANISMS

The fungus (*Ustilago tritici*) causing loose smut in wheat normally hibernates as mycelium within the wheat seed, vegetates systemically, and sporulates in the young wheat heads previous to their emergence, destroying most of the tissues of the spike, except the rachis, and producing a black powdery mass of smut spores in their stead. Smutted and healthy heads emerge during approximately the same period, and spores from the former are disseminated through the air while the healthy heads are in bloom. Inoculation occurs through the chance falling of one or more spores on the ovary or stigma, which is exposed through the opening of the glumes in the process of blooming. Within a few days all the spores are blown or washed from the diseased heads, and only the naked rachises of the spikes persist. Following their inclusion within the glumes of the healthy heads, the spores soon germinate. The germ tubes eventually enter the developing kernel and produce a mycelium, which hibernates within the matured kernel, thus completing the life cycle.

According to Freeman and Johnson (*6*), the life history of the loose-smut fungus of barley is similar to that of the loose-smut fungus of wheat. Like the latter, it infects its host only through the open flower and hibernates within the kernel as mycelium. To render the fungus innocuous it is necessary to apply a treatment like the modified hot-water method, which thoroughly soaks the seed. In recent years, however, Tisdale and Tapke (*13*) have produced loose smut in certain barleys as a result of seedling infection from smut spores applied to the mature seed, and Tisdale et al. (*14*), Tisdale and Tapke (*13*), Kirby (*7*), and others have noted that there is a varietal difference in the behavior of barleys toward seed treatment for the control of loose smut. Treatment of the seed with formaldehyde and certain organic mercury compounds has given partial or effective control in some varieties. Pending further addition to a knowledge of the factors involved in these new developments, the writer's studies reported herein have been confined to the floral infection of barley described by Freeman and Johnson (*6*).

PREVIOUS INVESTIGATIONS

Apparently, no previous investigations have been undertaken specifically to determine the rôle of humidity either in the infection, distribution, or control of the loose-smut fungi of wheat and barley, or any of the other cereals. However, several observations have been reported that have a bearing on the subject.

Mackie (*10, p. 618*) notes that "in California the dry season is near at hand when cereal plants are heading. Usually no considerable quantity of rain falls after this time," and that (*9, p. 2*) "the damage from loose smuts in wheat, barley, and oats in California is so slight that these smuts are not considered of economic importance." In studies on the floral-infecting loose smut of oats in Germany, Diehl (*5*) observed that smut spores alighting on the ovary of the blossoming flower germinate at once, except in very dry years.

PRESENT INVESTIGATIONS

EXPERIMENTS IN 1923–24

In the previous studies on the relative resistance of different wheat varieties, mostly eastern varieties, to loose smut (*12*), the writer included in the test in 1924 the three club varieties Hybrid 128 (C. I. 4229), Jenkin (C. I. 5177), and Little Club (C. I. 4066). These wheats are widely grown in one or more of the Pacific Coast States (*4*) where loose smut is very uncommon. (Fig. 1.) Smut-free plants from seed treated by the modified hot-water method were grown in a greenhouse at the Arlington Experiment Farm, Rosslyn, Va., in 1923. Flowers on these plants were inoculated by gently placing, with forceps, a small quantity of loose-smut spores on the stigmas of flowers in the process of blooming. The seed infected from such inoculation was harvested at maturity in March, 1923, and sown in a greenhouse the following autumn. The plants from this seeding were harvested in February, 1924, when fully headed. The number of plants and heads and the percentage of smutted plants and heads were obtained. The data are presented in Table 1.

TABLE 1.—*Percentages of loose smut in three varieties of club wheat grown in the greenhouse from seed from artificially inoculated flowers, Arlington Experiment Farm, Rosslyn, Va., 1923–24*

Variety	C. I. No.	Seeds sown	Plants matured		Plants smutted		Heads		
							Total	Smutted	
		Number	*Number*	*Per cent*	*Number*	*Per cent*	*Number*	*Number*	*Per cent*
Hybrid 128	4229	170	163	95.88	152	93.25	528	473	89.58
Jenkin	5177	140	138	98.57	134	97.10	463	452	97.62
Little Club	4066	175	160	91.42	156	97.50	651	632	97.08

Table 1 shows that the three club wheats, which rarely become smutted when grown in their habitat in the United States, were extremely susceptible when inoculated under the humid conditions prevailing in the greenhouse.

EXPERIMENTS IN 1927–28

In view of the suggestiveness of the data obtained in the foregoing preliminary experiment, the investigation was continued to determine definitely whether or not loose smut in wheat might be reduced or controlled by subjecting inoculated flowers to the low relative humidities generally prevalent during the blooming period of wheat in those parts of the United States where loose smut is uncommon.

MATERIALS, METHODS, AND RESULTS

Plants of a pure-line selection of Little Club (C. I. 4066) wheat were grown in 4-inch pots in a greenhouse at Arlington Experiment Farm, Rosslyn, Va., in 1927. At blooming, flowers were inoculated with freshly matured spores of *Ustilago tritici* from Little Club grown in another greenhouse. The technic of inoculation previously noted (p. 507) was again employed. Plants were inoculated on May 10, 1927, and placed under either arid or humid conditions for a period of eight days after inoculation. The plants subjected to humid conditions

were placed in a greenhouse in which the relative humidity varied between 56 and 85 per cent. The greenhouse floor was sprinkled from time to time to maintain a high humidity. Following this 8-day period, the plants were left to mature under the usual greenhouse conditions.

The arid condition was simulated by placing inoculated heads inside an elevated chamber containing air with a relative humidity reduced to 11 to 30 per cent through the use of calcium chloride. (Fig. 3.) The chamber was elevated so that the bottom was a few inches lower than the base of the heads of the shortest wheat plants.

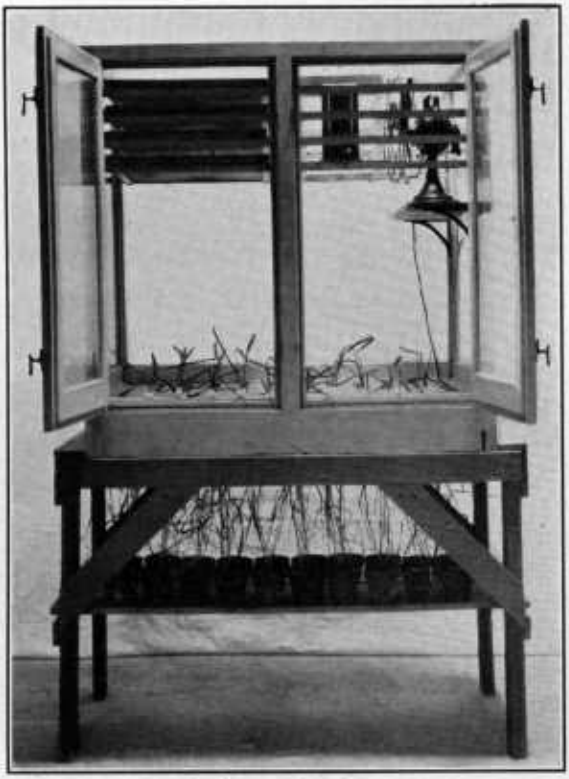

FIGURE 3.—Chamber used for subjecting wheat flowers inoculated with loose smut to low humidity

The bottom of the chamber contained numerous holes, seven-eighths of an inch in diameter, tightly plugged with corks. In order to introduce an inoculated head into the chamber, an opening was made by removing a cork. The head of wheat was inserted through the hole, as the plant bearing it was placed on the floor beneath the chamber. A ring of absorbent cotton was then tightly wound around the culm, at and just below the point of its passage through the bottom of the chamber, and tightly wadded into the hole. By this method inoculated heads were placed inside the chamber without noticeable effect on the moisture content of the contained air.

A low humidity was maintained by suspending 12 pounds of calcium chloride in shallow pans on racks near the roof of the chamber. An electric fan placed at one end of the pans circulated the contained air over the chemical. Two lots of calcium chloride were used. Each day one lot was removed from the chamber and replaced by the other, previously heated for three hours at 100° C. in a gas oven. The exchange was made at 10 a. m. daily. Following this operation, the relative humidity inside the chamber went down to 11 per cent within an hour, increasing to 30 per cent as the drying material became less absorptive.

Relative humidity was determined by a hygrometer placed in front of an electric fan as shown in Figure 3. The hygrometer was inserted, left 20 minutes, and read. It was then removed so as to influence the relative humidity inside the chamber as little as possible.

Flowering heads of Little Club (C. I. 4066) wheat were inoculated by hand with freshly matured chlamydospores of *Ustilago tritici* and then left for eight days either in a humid greenhouse or in the low-humidity chamber. Each day during incubation a record of the humidity and temperature was made at 9 a. m., 1 p. m., 3 p. m., and 5 p. m. Relative-humidity percentages in the arid and humid groups ranged from 11 to 30 per cent and from 56 to 85 per cent, respectively. Temperatures ranged from 70° to 92° and from 67° to 84° F., respectively. The low-humidity chamber was protected from the direct rays of the sun, but, nevertheless, on bright days it was warmer than the humid greenhouse. The plants were removed from the low-humidity chamber on May 18. Plants of both groups were then placed together in the greenhouse and left to mature. According to Freeman and Johnson (*6*), wheat becomes increasingly resistant to infection as the period after blooming lengthens, losing its susceptibility entirely by the time the ovaries are one-third developed. At the end of eight days the ovaries of the inoculated flowers were more than one-third of their mature size and had passed the stage of susceptibility to infection, regardless of subsequent changes in environment. Any inoculum remaining in the flowers after the 8-day period, therefore, was incapable of producing infection. Seedling infection of wheat by *U. tritici* resulting from spores on the mature seed apparently has never been obtained. The 8-day postinoculation period also provided ample time for the fungus to reach its ultimate objective, the cavity of the ovary. According to Lang (*8*), this is normally accomplished in about one week after inoculation.

At maturity the kernels, developed in the inoculated flowers, were harvested and stored until the following winter (1927–28), when they were sown in the greenhouse. The resulting crop was harvested at maturity in the early spring of 1928. The results are presented in Table 2 and illustrated in Figures 4 and 5.

TABLE 2.—*Percentages of loose smut in Little Club wheat (C. I. 4066), grown in the greenhouse from seed from flowers incubated under the given conditions of humidity and temperature for eight days after artificial inoculation, Arlington Experiment Farm, Rosslyn, Va., 1927–28*

Atmospheric conditions during incubation		Seeds sown	Plants matured		Plants smutted		Heads		
Range in relative humidity	Range in temperature						Total	Smutted	
Per cent	° *F.*	*Number*	*Number*	*Per cent*	*Number*	*Per cent*	*Number*	*Number*	*Per cent*
56–85	67–84	140	107	76.43	102	95.33	182	171	93.96
11–30	70–92	275	199	72.36	43	21.61	315	69	21.90

FIGURE 4.—Loose smut in Little Club wheat grown from seed from inoculated flowers subjected to a range of relative humidity of 56 to 85 per cent for eight days after inoculation. A, healthy plants; B, smutted plants

Table 2 and Figures 4 and 5 show a striking reduction in smut infection when inoculated flowers were subjected to ranges of low relative humidity and temperature similar to those occurring in the areas of the United States free from loose smut (fig. 1) during the blossoming period of wheat.

The influence of the difference in the ranges of temperature in this experiment can hardly be evaluated. According to Appel and Riehm (*1*), the cardinal temperatures for the germination of loose-smut spores are 42.8° to 50°, 80.6° and 91.4° to 93.2° F. Spores also were found capable of germination after an exposure of 12 hours to a temperature

of 96.8° In this experiment the temperatures were all within the germination range. It seems likely, therefore, that differences in humidity probably were accountable for most of the difference in loose-smut infection.

After the inoculations on May 10, several inoculated flowers were taken each day from plants in each of the two groups and examined microscopically. In the group held under humid conditions many spores were in the early stages of germination one day after inoculation. On the second and third days after inoculation approximately 90 per cent of the spores had germinated and produced long germ tubes intermingled with and ramifying the stigmas. The germina-

FIGURE 5.—Loose smut in Little Club wheat grown from seed from inoculated flowers subjected to a range of relative humidity of 11 to 30 per cent for eight days after inoculation. A, healthy plants; B, smutted plants

tion of the spores in flowers of the group held at low humidities presented a wholly different situation. Spores in the early stages of germination were not found until the third day after inoculation, and these were few in number. In later examinations, up to and including the time when the inoculated heads were removed from the dry chamber, only a few spores were found with germ tubes as long as those produced by spores in the other group two and three days after inoculation. These observations would seem to indicate that in the presence of low humidity loose smut in wheat is held in check because the smut spores either fail to germinate or germinate too slowly to enable the infecting hyphae to reach the ovary during the short period in which it is vulnerable.

EXPERIMENTS IN 1928–29

The study was continued at the Aberdeen substation, Aberdeen, Idaho, in 1928. In this year the experiments included the loose smut of barley in addition to the loose smut of wheat. Humid conditions were obtained by covering and spraying plants growing on irrigated land. Arid conditions were obtained by placing plants, in containers, in the desert and exposing them to the normally dry air of this arid region.

MATERIALS, METHODS, AND RESULTS

Federation (C. I. 4734) wheat and Moultan (C. I. 3401) and Hanna (C. I. 203) barleys were used. Federation wheat and Hanna barley were inoculated at the beginning of the second week in July, and Moultan barley was inoculated at the end of the third week in July. The technic of inoculation described on page 507 was employed. The inoculum used on Federation wheat consisted of chlamydospores of *Ustilago tritici* collected from the same variety a few days previous to inoculation. The smut used to inoculate the barleys consisted of a mixture of chlamydospores of *U. nuda* collected shortly before inoculation from different varieties of barley growing on the farm of the Aberdeen substation.

FIGURE 6.—Type of container used for subjecting inoculated wheat and barley flowers to high humidity on irrigated land

In the group subjected to a humid atmosphere after blossom inoculation, enough flowers of one of the given varieties were inoculated during one day to serve the purpose of the test. Flowers on plants growing in a clump in a row or plot on the irrigated land were inoculated. The plants of the high-humidity group were placed in chambers made of wall board and were sprayed with water twice daily for eight days after inoculation. The doors of the chambers were closed during the first three days and opened (fig. 6) during the remainder of the 8-day treatment. On the ninth day after inoculation the chambers were removed, and the plants were left to mature. Humidity and temperature records taken daily at 9 a. m., 12 m., and 5 p. m. during the 8-day period showed ranges for Federation wheat and Hanna barley of 45 to 92 per cent and 58° to 89° F., and for Moultan barley of 42 to 89 per cent and 67° to 91°. During the 3-day postinoculation period in which the doors of the chambers were closed, the relative humidity of the contained atmosphere was generally above 75 per cent.

Wheat and barley plants subjected to arid conditions were grown on the irrigated farm until about two weeks before heading. Clumps of the plants were then carefully dug and placed in galvanized-metal cans. The plants suffered no apparent injury from transplanting when the clumps were removed from the field and carefully placed in the cans so that the ball of earth incasing the roots was neither cracked nor broken. At the beginning of blossoming, the flowers were inoculated, and the plants were placed in a finely screened cage in a desert field. The cage was located about 500 yards to the windward of the irrigated plots of the experiment farm. (Fig. 7.) Test readings taken at midday on a clear typical day early in July showed a relative humidity of 13 per cent, both in the cage and at a spot in the desert 3 miles to the windward of the nearest irrigated farm. Records taken at 9 a. m., 12 m., and 5 p. m. during the 8-day period following inoculation showed humidity and temperature ranges for Federation wheat and Hanna barley of 13 to 48 per cent and 53° to 95° F., and for Moultan barley of 12 to 30 per cent and 62° to 98° F. Following inoculation the plants of this group were left in the desert until mature.

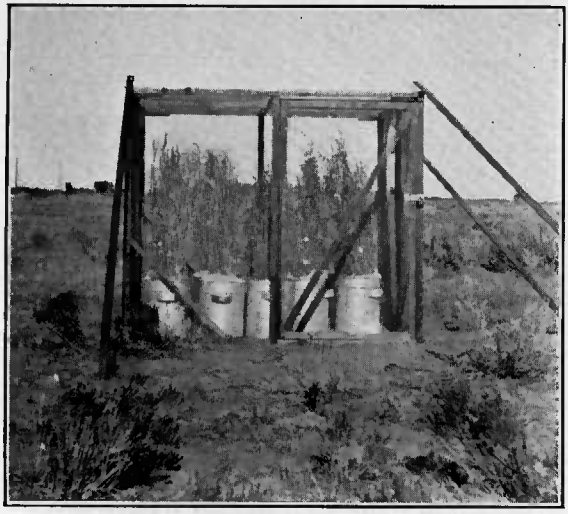

FIGURE 7.—Apparatus used in subjecting inoculated wheat and barley flowers to the low humidity of the desert

Seed of both groups was harvested at maturity and stored in sealed glass jars. The following November it was sown in a greenhouse at Arlington Experiment Farm, Rosslyn, Va. Records on smut infection were taken after the plants were fully headed. The data are presented in Table 3.

Plants from seed developed from inoculated flowers incubated in the arid atmosphere of the desert produced much less loose smut than plants from seed developed from inoculated flowers incubated in the humid atmosphere on the irrigated land. The reduction in infection in plants of Moultan barley was especially striking (83.93 to 0 per cent). The lower humidity and higher temperature prevailing during the period in which Moultan was inoculated (Table 3) probably may be held accountable for the more effective control of smut in this variety.

In this experiment, as in the greenhouse experiment of 1927–28, there were differences in temperature as well as in humidity in the arid and humid groups. It is difficult or impossible to evaluate the influence of such differences in temperature range. Further work is necessary with apparatus that will secure variations in relative humidity with a constant temperature.

TABLE 3.—*Percentages of loose smut in Federation wheat and Hanna and Moultan barleys grown in the greenhouse from seed from flowers incubated under the given conditions of humidity and temperature for eight days after artificial inoculation at Aberdeen, Idaho, July 1, 1928*

IN HIGH-HUMIDITY CHAMBERS ON IRRIGATED LAND

Variety and C. I. No.	Atmospheric conditions during incubation		Plants			Heads		
	Range in relative humidity	Range in temperature	Total	Smutted		Total	Smutted	
	Per cent	*° F.*	*Number*	*Number*	*Per cent*	*Number*	*Number*	*Per cent*
Federation wheat (C. I. 4734)	45–92	58–89	166	116	69.88	240	164	68.33
Hanna barley (C. I. 203)	45–92	58–89	45	28	62.22	142	81	57.04
Moultan barley (C. I. 3401)	42–89	67–91	70	61	87.14	168	141	83.93
UNDER DESERT CONDITIONS								
Federation wheat (C. I. 4734)	13–48	53–95	203	37	18.23	341	49	14.37
Hanna barley (C. I. 203)	13–48	53–95	81	12	14.81	264	27	10.23
Moultan barley (C. I. 3401)	12–30	62–98	108	0	0	257	0	0

DISCUSSION

The data show that low relative humidity during the period of blossoming plays an important rôle in preventing the infection of wheat and barley with their respective loose smuts. By growing a crop in a dry area for one year it would seem possible to rid it of loose smut without seed treatment. This might have a practical use worthy of some consideration in view of the fact that the heavy annual loss from loose smut in wheat and barley goes on unchecked largely because the seed treatment recommended for control is difficult to apply and is likely to reduce germination and yield. Clean seed might be produced in arid areas either under dry farming or by proper manipulation of irrigation. The important consideration is low humidity during blossoming.

The knowledge of the effect of low atmospheric humidity on the infection of wheat by *Ustilago tritici* would seem to be particularly applicable in the case of highly susceptible varieties grown under irrigation, such as Dicklow and Federation. Dicklow is extensively grown under irrigation in southern Idaho and to a lesser extent in Montana, Utah, and Wyoming (*4*). Federation is now widely grown under irrigation in Idaho. A modification of irrigation practice to induce a low humidity during blossoming should reduce infection. The details should be determined by experiment.

SUMMARY AND CONCLUSIONS

Loose smut in wheat and barley rarely occurs in the Pacific Coast States and in other sections of the country where the relative humidity of the air is low at the flowering time of these cereals. It is at this time that the loose-smut spores are blown from diseased heads into the open blossoms, where normally they germinate and produce infection hyphae that enter the developing kernel.

When flowers of the three club wheats Hybrid 128, Jenkin, and Little Club were inoculated under humid conditions in a greenhouse at the Arlington Experiment Farm, Rosslyn, Va., the resulting seed produced from 91 to 98 per cent smutted plants. These wheats are widely grown in one or more of the Pacific Coast States and rarely produce loose smut there.

Flowers of Little Club wheat were exposed for eight days after inoculation to low (11 to 30 per cent) and high (56 to 85 per cent) ranges in relative humidity. Seed from flowers subjected to the low humidity produced 21.90 per cent smutted plants, while seed from flowers subjected to the high humidity produced 93.96 per cent smutted plants. In this experiment the low humidity was obtained by recirculating air over calcium chloride in a closed chamber.

Daily microscopic examinations of inoculated flowers showed that in plants exposed to humid conditions many spores were in the early stages of germination one day after inoculation, and that on the third day following inoculation approximately 90 per cent of the spores had germinated and produced long germ tubes. In the group exposed to arid conditions, spores in the early stages of germination were not found until the third day after inoculation, and only a few with long germ tubes were found seven days after inoculation. These observations seem to indicate that in the presence of appropriately low relative humidity loose smut in wheat is held in check because the smut spores either fail to germinate or germinate too slowly to enable infecting hyphae to reach the ovary during the short period in which it is vulnerable.

At Aberdeen, Idaho, flowers of Federation wheat and of Moultan and Hanna barleys were inoculated on plants on the desert and on plants on irrigated land. In each variety the seed from flowers inoculated and incubated under desert conditions produced a much lower percentage of smutted plants and heads than the seed from flowers inoculated and incubated under irrigation conditions. Plants from seed from the inoculated flowers of Moultan barley incubated under high humidity produced 83.93 per cent of smutted plants as compared with 0 per cent of smutted plants from seed from inoculated flowers incubated at a low humidity.

In the experiments involving a comparison of the effects on infection of high or low ranges of humidity, the latter were accompanied by ranges of temperature greater than those accompanying the high humidity ranges. However, the ranges of temperature did not extend beyond the limits for germination of the spores of *Ustilago tritici* and *U. nuda* except in the case of Moultan barley. The possible influence of these temperature differences must be recognized, but they can not be evaluated in these experiments.

The data suggest a practical method for producing seed stocks free from loose smut by growing them under conditions of low relative humidity during the blossoming period.

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