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BIOLOGICAL DEPARTMENT
CHEMICAL CORPS

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BIOLOGICAL DEPARTMENT, CHEMICAL CORPS
SO and C DIVISIONS
Camp Detrick, Frederick, Maryland

SPECIAL REPORT NO. 138

15 December 1950

29 Aug 1977
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FEATHERS AS CARRIERS OF BIOLOGICAL WARFARE AGENTS

I. Cereal Rust Spores

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APPROVED:

Oram C. Woolpert
ORAM C. WOOLPERT, N.D.
Director
Biological Department

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INTRODUCTION

It has been demonstrated that birds dusted with cereal rust spores will retain sufficient numbers of spores to initiate a cereal rust infection.

The first test, conducted at Camp Detrick, consisted of dusting birds with cereal rust spores (Puccinia graminis avenae, Race 8) and releasing them for 1 1/2 to 2 1/2 hours in cages covering approximately 100 square feet of seedling Vicland oats. A heavy rust infection resulted on all of the plots.

A second test was conducted to determine if birds dusted with rust spores will retain sufficient numbers of the spores to cause infection after a 100-mile flight. Ten homing pigeons dusted with rust spores were released and allowed to fly approximately 100 miles to their home barn. Four of these birds were placed for 2 hours in a cage covering approximately 50 square feet of seedling Vicland oats. Sample feathers were removed from 2 of the 4 pigeons at intervals over a 19-day period. This test demonstrated that sufficient spores will be retained on birds after a 100-mile free flight to initiate primary infection; it was also shown that large numbers of viable spores will remain on these birds for at least 19 days.

In the third test, conducted at St. Thomas, Virgin Islands, four test plots covering 1600 square feet each of Vicland oats were prepared approximately 1/2 mile apart on an isolated island. Four groups of pigeons, trained to return to their respective plots, were dusted with rust spores and liberated from an airplane. The birds returning to the plots were allowed to remain on the plots for approximately 2 hours. Heavy infection resulted in all plots, demonstrating that birds dusted with rust spores and released from aircraft will retain sufficient numbers of spores to initiate a cereal rust infection.

These tests demonstrated that birds dusted with rust spores can carry these spores to cereal plants where rust infection may result under certain conditions. The problems involved in collecting, stock piling, and processing birds would tend to limit the use of this method for the wide-scale distribution of cereal rust spores over an enemy territory. However, these studies did show that large numbers of viable spores remain attached to the feathers of birds for considerable periods. On the basis of this observation, it appeared that feathers alone possibly could be used as carriers of BW materials.

The purpose of the feather test described in this report was to determine if feathers dusted with cereal rust spores and released from aircraft in an M16A1 cluster adapter (used for leaflets and fragmentation bombs) will permit the transference of spores to cereal plants so

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that rust infection may ensue. In addition, undusted feathers were released at several altitudes to determine the distribution pattern on the ground.

Acknowledgements: The facilities for this test were made available to the Chemical Corps by the Commanding General, 1st Army, and by the Commanding Officer, Pine Camp, New York.

Lt. Col. Robert C. Edgar, Post Engineer, Pine Camp, New York, was instrumental in supplying the necessary equipment and vehicles required to conduct the test.

Captain Glenn E. Davis, Captain George Nielsen, Major Horace A. Templeton, and Lt. Raymond E. Horne, Air Force Field Office, Camp Detrick, Maryland, aided materially in locating and preparing the test area and in conducting these trials.

MATERIALS AND METHODS

The munition used to release the feathers at the desired altitude was a modified M16A1 cluster adapter equipped with a MT,AN-M146 bomb nose fuse. This standard cluster adapter was modified (Figure 1) so that the packages containing the feathers would open and the contents would be discharged upon the functioning of the cluster. Two straps (1/2 in. metal strapping) were attached on the inner surface of each package, one strap being attached to the upper section of the cluster and the other to the lower.

The feathers used for this test were washed, fluffed, white turkey plumage of a uniform size (average 3 1/2 in. long by 1 3/4 in. wide; 5300 per pound). (Figure 2)

Such feathers will hold loosely up to 80 per cent by weight of rust spores. After violent shaking approximately 10 per cent by weight of spores are retained. The high retentive characteristics of the feather are due to its anatomy (Figure 2) and to the structure of the spore (Figure 3). The feather contains numerous barbs and hooklets which act as pockets to retain the spores which are likewise barbed. For this test, 1 part spores to 10 parts of feathers by weight were used, but the optimum spore/feather ratio needs to be determined.

Puccinia graminis avenae, Race 8, a parasitic fungus of oats, was the test agent. The uredospores (Figure 3) of this agent were produced in field plots and collected with the spore harvester described in C Division Report for the Third Quarter, 1949.

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The feathers and rust spores were mixed by placing 2 1/2 pounds of feathers and 1/4 pound of rust spores into a mixing drum (Figure 4). The drum was rotated at 9 rpm for 30 minutes. The dusted feathers (Figures 5 and 6) were hermetically sealed in a polyethylene-coated kraft paper package (2 mil, 30 lb weight) and stored at 36° F until used. Four such packages were used per cluster.

The target area consisted of sixteen 1/2-acre plots planted with Overland variety oats, which is susceptible only to race 8 of Puccinia graminis avenae and is resistant to other common races of stem rust and to crown rust. These plots were located in a rectangular area approximately 11 miles long by 1 1/2 miles wide (Figure 7). The oats had been seeded 6 weeks prior to the test and ranged in stage of development from late tillering to early boot.

PROCEDURE

Biological Test: Three clusters loaded with dusted feathers as described above were released approximately one mile upwind of the target area. Two were opened at 1300 feet above ground level and one at 1800 feet. A 15- to 27-mile per hour wind (Table I) distributed the feathers over 7.7 square miles (Figure 8). Due to the meteorological conditions at the time of the test, feathers fell on only 5 of the 16 plots. Over 100 feathers were found in each of 2 plots, while single feathers were found in each of the other 3 plots. Observers located near the plots noted the number and the location of feathers landing on each plot.

The experimental plots were observed for extent and severity of infection during a period of 40 days following the distribution of the dusted feathers. The experimental planting was destroyed immediately after final observations were made.

The daily maximum and minimum temperatures and precipitation for the period of the test are shown in Table II.

Distribution Studies: Six cluster adapters each containing 10 pounds of feathers (identified by different colors) were released and set to open at the following altitudes above ground level: 1300, 1800, 2000, 2300, 3000, and 4000 feet. An observation plane circled the clouds and indicated to ground observers the general areas of distribution. Six teams of observers, each assigned to a particular area, recorded the distribution patterns of the different colored feathers.

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RESULTS

Biological: Two weeks after the release of the munition, stem rust was found in 11 of the plots located nearest the impact area (Figure 9). Rust pustules were at about the same stage of development on oat plants in these plots and on plants artificially inoculated at the time the feathers reached the plots. More than 100 feathers were found on each of 2 plots and single feathers on 3 more. No feathers were found on the remaining 6 of the 11 plots showing primary infection. Primary rust infection on the plots on which feathers fell was not greater than on plots in the vicinity which had escaped direct contact with feathers.

Detailed observations on rust infection were begun 2 weeks after the liberation of the spore-dusted feathers and continued for 4 weeks. Information was obtained on the proportionate number of plants infected (prevalence) and on the estimated percentage of plant area covered with rust pustules (severity). Primary infection on the 11 plots attacked initially ranged from 2 to 15 per cent prevalence (Table III) with a severity of less than 0.1 per cent (Figure 11A). Three weeks later all plants in these plots were infected by rust and the severity had increased to 7 to 30 per cent (Figure 11B). Rust continued to increase during the last week of the trial.

The 5 plots most distant from the impact area which were not initially infected, developed rust between 1 and 2 weeks following the appearance of rust spores on the other 11 plots. This secondary spread apparently resulted from wind-blown spores (Figure 9).

It was necessary to discontinue the test before the oat crop matured, because of the onset of fall weather. Actual losses in grain resulting from the rust attack could not be determined, but correlations between rust severity and stage of plant development have been used with reasonable accuracy for many years in estimating crop losses. Such correlations indicate that loss in grain on the 11 plots first attacked would have been at least 30 per cent, while the remaining 5 plots were also severely injured.

Analysis of oat stem rust samples obtained from farmers' oat fields and from volunteer oat plants in the vicinity of Pine Camp prior to and during the test showed that the predominant race was No. 7. Race 8, which was used as the experimental agent, was found only in the test plots during the test and on collections from volunteer grain towards the end of the test period.

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Distribution Trials: The results of the 6 drop-tests made to determine the areas of distribution at different altitudes are summarized as follows:

<u>Opening Altitude</u> <u>(Approx. Ft.)</u>	<u>Average Wind Speed</u> <u>(Miles per Hour)</u>	<u>Areas of Distribution</u> <u>(Square Miles)</u>
1300	25	4.6
1800	25	5.0
2000	23	4.6
2300	25	6.5
3000	23	8.2
4000	23	12.5

These results are shown graphically in Figures 12 through 17.

DISCUSSION

The test described herein was conducted to determine whether rust infection would occur on cereal plants following the release of spore-dusted feathers from an M16A1 cluster adapter released from an airplane. Rust infection developed on cereal plants following such distribution of dusted feathers.

Rust was found on 11 of the plots nearest the impact area 2 weeks after the liberation of the dusted feathers. Feathers had fallen only on 5 of the 11 plots on which infected plants were found initially. Spores separated from feathers were, no doubt, responsible for much of the infection; this is shown by the following: the 5 plots where feathers were found had infections near carriers as well as some distance away and the remaining 6 plots also developed cereal rust. In those plots where feathers had fallen about 10 per cent of the plants were infected with a few scattered pustules. Occasionally 1 plant or more rarely several plants in a group were infected with many pustules. Plants showing many pustules had the type of rust pustule distribution seen when a spore-dusted feather was rubbed on leaves and stems. The scattered infection of a few pustules per infected plant was characteristic of infection following the deposit of free spores.

The relatively heavy primary infection occurring on a plot located 2 1/2 miles upwind of the impact area must certainly have resulted from free spores, since dusted feathers were not found within 2 1/2 miles of this plot. A ground wind shift of 180° that occurred 24 to 30 hours

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following the liberation of the feathers presumably was responsible for carrying the spores to this plot.

Spore distribution following the release of the feathers was estimated to be about 25 square miles, this figure being calculated from the distribution of primary rust pustules on the experimental oat plantings. The feathers carrying the rust spores were spread over 7.7 square miles, thus, the spore distribution was at least 3 times that of the feathers in this test.

Trials with other lots of feathers of uniform size and at relatively high wind velocities showed that feathers were dispersed over from 4.6 square miles when liberated at 1300 feet to 12.5 square miles when liberated at 4000 square feet. Additional trials with feathers of different sizes should be conducted to evaluate the dispersal pattern obtained by feather liberation at several heights and at a number of wind speeds.

Accurate data on the loss of grain resulting from the rust infection could not be obtained, but correlations between severity of rust and stage of host plant development indicated that the yield on the plots primarily infected was reduced by at least 30 per cent.

CONCLUSIONS

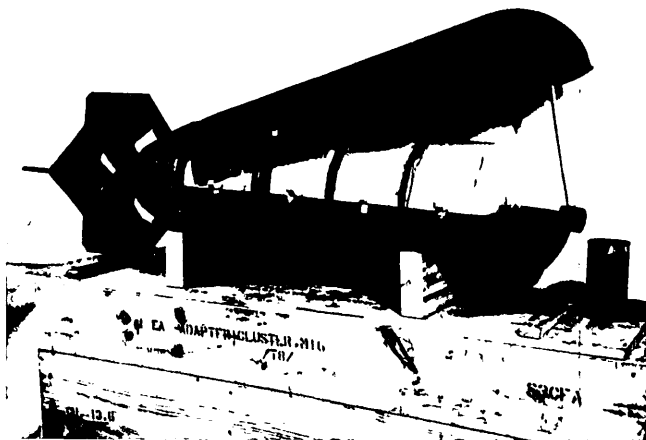
It is concluded that feathers dusted with 10 per cent by weight of cereal rust spores and released from a modified M16A1 cluster adapter at 1300 to 1800 feet above ground level will carry sufficient numbers of spores to initiate a cereal rust epidemic.

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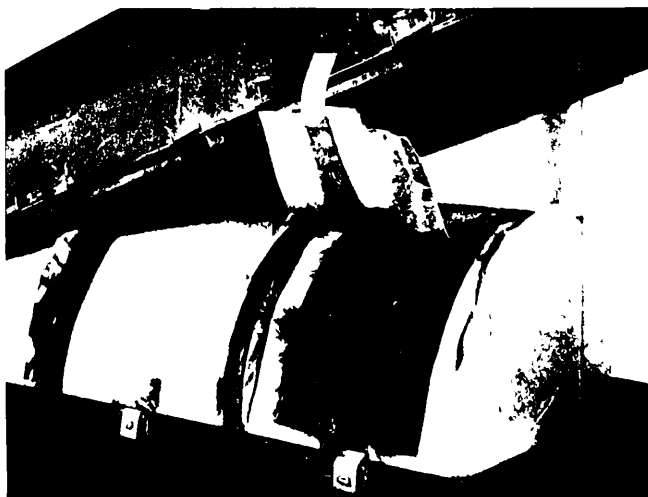
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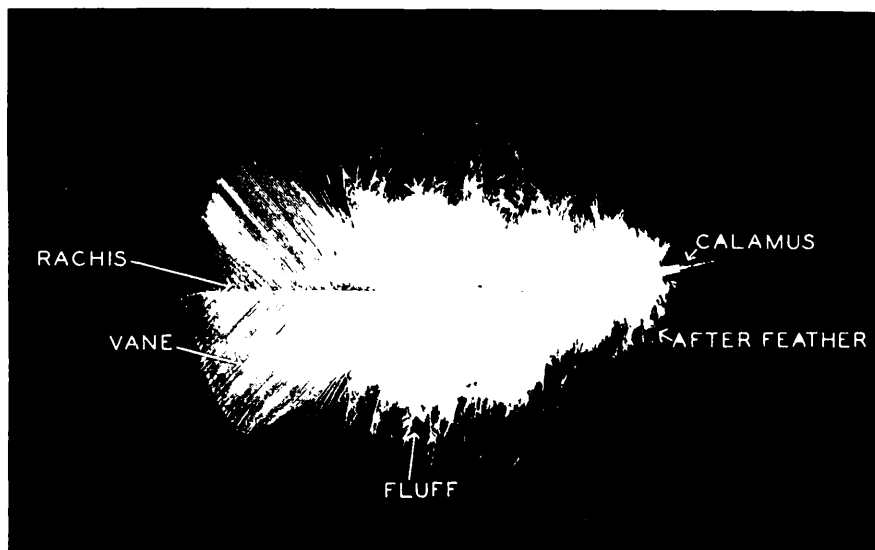


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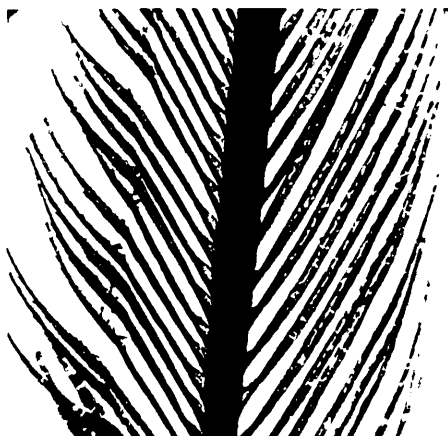
FIGURE 1 (A) MODIFIED CLUSTER ADAPTER
M16 (B) CUTAWAY SECTION OF
PACKAGE

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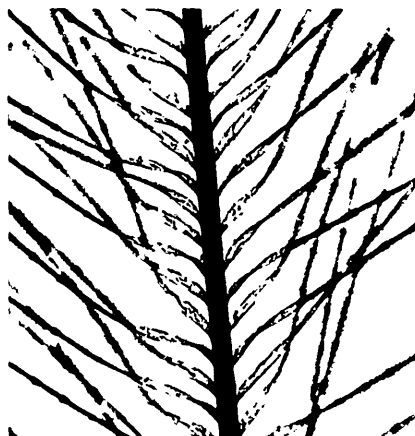
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NATURAL SIZE



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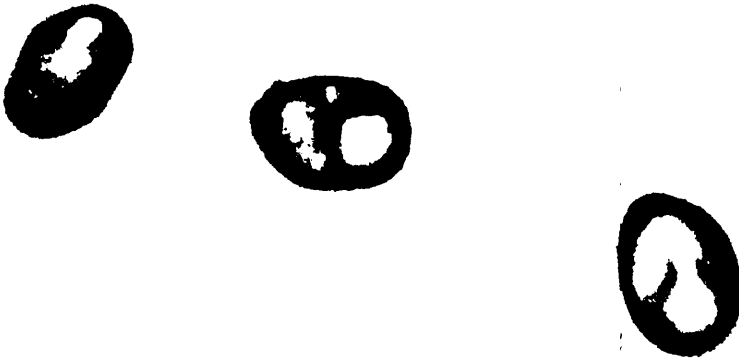


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FIGURE 2 ANATOMY OF TURKEY FEATHER

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**FIGURE 3A PHOTOMICROGRAPH OF STEM
RUST SPORES (APPROX x 1000)**



**FIGURE 3B ELECTRON MICROGRAPH OF
STEM RUST SPORE (x 15,000)**

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