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DIVISION OF PLANT PATHOLOGY AND PHYSIOLOGY

A STUDY OF THE BLACK AND THE YELLOW MOLDS OF EAR CORN

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A STUDY OF THE BLACK AND THE YELLOW MOLDS OF EAR CORN.

BY J. J. TAUBENHAUS.

The present bulletin is the result of four years' careful study of the black and the yellow molds of ear corn. These studies were undertaken at the insistent request of corn growers in Texas. That these diseases are of considerable economic importance may be seen from the following figures.

A three-year average of the Texas corn crop for 1917, 1918, and 1919, was estimated by the Monthly Crop Reporter* as 114,566,666 bushels, with an average price of \$176,432,666.66. A conservative estimate of the average annual loss from the black and the yellow molds of ear corn during that time is 5,718,333 bushels. If the average price for the last three years is taken to be \$1.53 per bushel, then the average annual money loss from the ear molds of corn was \$8,806,233. Since the black and the yellow molds of ear corn also attack broom corn, it would not be out of place to carry the figures a step further. A three-year average of the production of broom corn for 1917, 1918, and 1919 is figured at 12,800 tons with an average price of \$305,933. In estimating the losses from the black and the yellow molds of broom corn at 4 per cent. of the total crop, there will be a loss of 512 tons, worth \$12,237. When the average annual loss of broom corn due to the presence of these molds is added to the average annual loss of field corn from the same cause, it may be asserted that the Texas growers are sustaining a yearly loss of \$8,818,349. The thoughtful farmer will at once realize the importance of being able to save this unnecessary waste. It should be added that as far as the corn crop is concerned, the greatest losses are sustained primarily from the black mold.

HISTORICAL.

In considering the economic importance of the black and the yellow molds of ear corn and broom corn in the Southern States, it is surprising to see how little mention is made in the literature about these diseases. Some entomologists early recognized the economic importance of these molds. Garman and Jewett (12) in 1914, in working with the corn-ear worm and in mentioning the extent of injury from this pest, state that seasons when corn mold is prevalent are merely seasons in which the insects have been especially common. Similarly, Bishop (3), in 1917, while working on the boll worm, or corn-ear worm, stated that in some regions, practically every ear of sweet corn was damaged, and that throughout the entire country 70 to 90 per cent.

*Monthly Crop Reporter, U. S. Department of Agriculture, 5:121-140, Dec. 1919.

of the ears of field corn were attacked. Following this injury, molds frequently gain access to the ears and damage them still further. This is especially true during wet seasons. Although the specific nature of the molds is not stated, it is reasonable to infer that Bishop was dealing with the black and the yellow molds, which are widespread in most of the Southern States. The great importance is the association of the ear-worm with the ear molds, a condition as is shown on page 7 that practically prevails in Texas.

As will be seen later, the cause of the black and the yellow molds of ear corn and of broom corn is due to two fungi belonging to the genus *Aspergillus*, namely: *A. niger* and *A. flavus*. Not only is *A. niger*, as determined by us, involved in the black mold of ear corn and broom corn, but as stated by Wayne (37 and 38), this fungus is responsible for serious damage to stored onions in Ohio. The same condition also prevails in Texas, especially in the Rio Grande Valley, where the onion crop is seriously damaged by *A. niger*. The black mold damages the crop more seriously when the onions are dug during wet weather, also making them more unfit for shipping. The symptoms of black mold of onions will be taken up on p. 18. Besides the onion, McMurrin (24) reports an internal rot of pomegranates which he attributes to a fungus, *Sterigmatocystis castanea* Patterson. As will be shown later, no distinction is made in this bulletin between *Sterigmatocystis* and *Aspergillus* as they are considered identical. Hodgson (18, 19, and 20) also records a pomegranate rot which he attributes to *Sterigmatocystis castanea* and a smut of figs also believed to be induced by the same fungus. Stevenson (33), while studying the cause of the rotting of citrus fruit in Porto Rico, found that among the many decay-producing organisms, *Aspergillus niger* was an important factor. Brooks and his colleagues (5) found that among the many rot-producing fungi, *A. niger*, was able seriously to injure ripe apples both on the market and in storage. Beille (2) found that *Aspergillus niger* and *A. flavus* were both involved in a stinking rot of coffee grains. Stoykowitch and Brocqrouseu (34) found that *Aspergillus* and *Penicillium* were responsible for the deterioration of dried prunes by reducing the sugar and acid content. The Kopeloffs (21 and 22) found that among the many fungi, *Aspergillus niger* was important in the deterioration of cane sugar. It should not be supposed that all species of *Aspergillus* are harmful. Wehmer (40) records that *Aspergillus oryzae* is very important in the brewing industry of the Japanese saki. Likewise, Hanzawa (15) states that *Aspergillus oryzae* is the chief fermentative agent in the manufacture of the Tamari-Kojii, a Japanese sauce made of soy beans.

PRESENT WORK.

The importance of the black and the yellow molds of ear corn and of broom corn in Texas has already been indicated on page 3. In the present work, an attempt has been made to determine the following:

- (1) The distribution of both the black and the yellow molds in Texas.
- (2) The exact relationship of *Aspergillus niger* and *A. flavus* to the molds of ear corn and broom corn.
- (3) The range of hosts of these two species of fungi.
- (4) The presence, if any, of physiological species.
- (5) The pathological effect of black mold on the germination of grains from affected ear corn.

(6) The morphology and physiology of *A. niger* and *A. flavus*.

(7) Methods of control.

The writer wishes to express his indebtedness to Dr. Charles Thom for many courtesies in verifying and identifying species of *Aspergillus*. Acknowledgment is also due Messrs. A. B. Conner and A. H. Leidigh of the Division of Agronomy, to Mr. H. B. Parks, of the Division of Entomology, and to Mr. A. S. Ware, secretary, for helpful criticism in reading the manuscript.

DISTRIBUTION OF EAR MOLDS.

As far as the ear molds of corn are concerned, it is necessary to distinguish between the black and the yellow mold. The term black mold or yellow mold is here used in preference to smut, as the disease is now erroneously known. The black mold, especially, must be distinguished from the true smuts of corn and broom corn in order to gain a clear conception of the difference in methods of treating these diseases. The black mold of ear corn may be generally found wherever corn is grown in Texas. As will be shown on page 24, this mold is more prevalent during dry seasons, although it is not uncommon during wet weather. The yellow mold, on the other hand, seems more restricted in its distribution and is especially prevalent during wet seasons. It should be emphasized in this connection that where yellow mold is common, black mold of ear corn is practically unimportant. As to the broom corn, both the black and the yellow molds usually occur together in the fields, being more prevalent on some varieties than on others. The greatest amount of damage to broom corn, however, is met with when the crop is harvested during wet weather and while green, when shipped before being thoroughly cured, or when loaded in cars which lack the proper amount of ventilation.

SYMPTOMS.

As the names indicate, the black and the yellow molds of ear corn attack only the ear and no other part of the corn plant. In the field there are practically no symptoms to indicate the presence of black moldy ears, inasmuch as the disease is confined to the interior of the ear itself and not to the exterior of the husks. It is only during harvesting and husking that the disease becomes apparent. The same is also true for the yellow mold, which is confined to the interior of the tip of the ear only (Fig. 3, a and b), in which case the infected grains are undersized, shriveled, and covered by a yellow growth which, if this growth is closely examined, it is found to be made up of numerous yellow-to-greenish heads, which are really the spore sacs of the fungus *Aspergillus flavus*. Yellow mold in the field seldom invades more than one-third or less of the tip of the ear. In this case it is nearly always found on varieties with erect ears, which catch and hold water from rain or dew. With the black mold, however, the disease is seldom confined to the tip, but follows any opening made by the ear worm (Figure 1, a, b, f, g, h, i, and j). In tearing open the husk of an infected ear one finds its entire surface is covered with a black powder, giving it a dark, sooty appearance (Figure 2, a, c, d, and e). The grains of such an ear are black, undersized, and in most cases shriveled and blackened (Figure 1, i). Invariably, and on close examination, it is found that infection

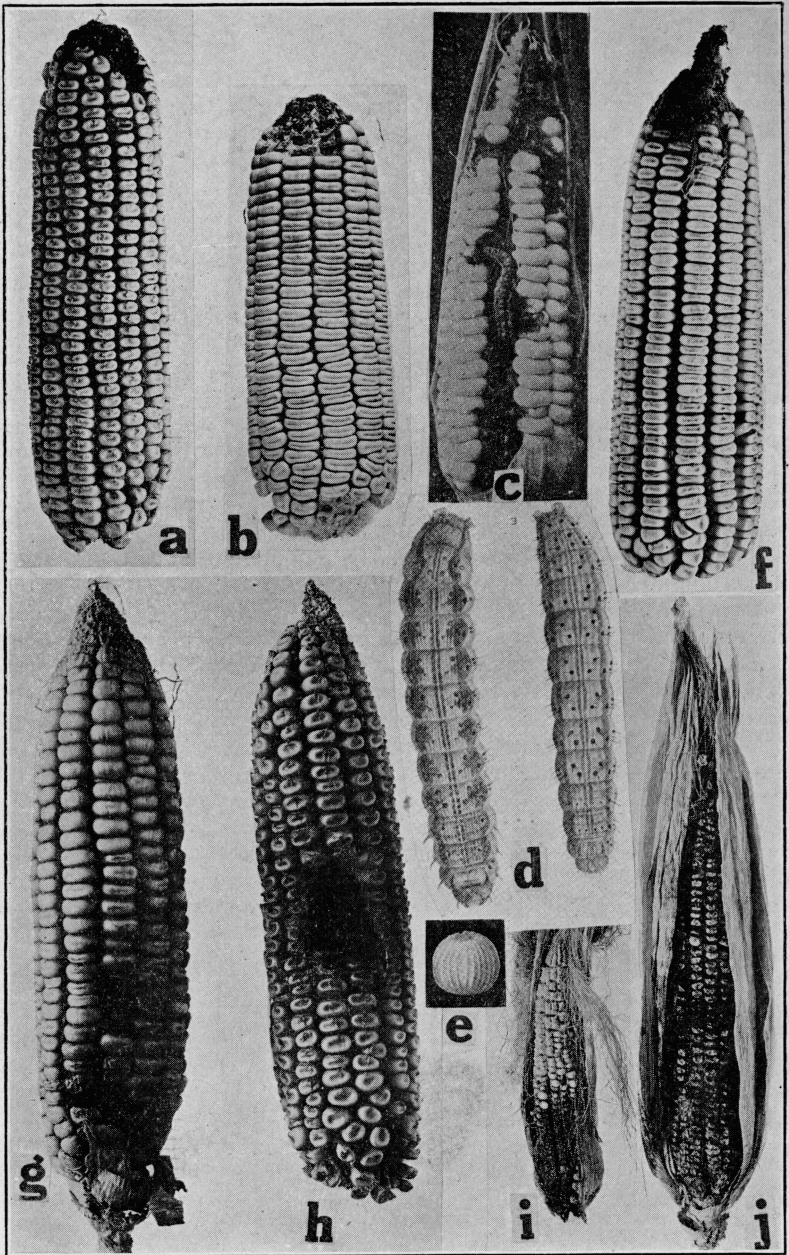


FIGURE I.

a. and *b.* Black mold starting at tip of ear and following ear worm injury. *b.* Same as *a.* except that the grains on the tip of the ear were removed to show that the black mold had penetrated the cob. *c.* Ear corn injured by the ear worm showing also the tunnels made by it. *d.* Two corn-ear worms. *e.* Eggs of corn-ear worm caterpillar. (*c.* After Quaintance and Bruce. *d.* and *e.* After Garman and Jewett.) *g.* and *h.* Black mold of ear corn starting in the middle and following the opening made by ear worm. *i.* and *j.* Two young ears of corn injured by ear worm and then destroyed by black mold.

of the black mold starts at the point where insects, especially the corn ear worm (Figure 1, d and e), have penetrated through the husk into the ear. The ear worm in feeding (Figure 1, c) produces considerable excrement, which is generally moist, thus offering an ideal medium for the entrance of *Aspergillus niger*. In early infections, the blackest part of the ear may be localized at or near the place of the worm injury. If the affected ear is young, tender, and in its milky stage, the fungus *A. niger* will be seen to spread rapidly from the point of entrance of the ear worm to the entire inner surface. When, however, the ear is partly ripe, infection will remain localized within the feeding area only of the ear worm. Infection seldom takes place on thoroughly ripened ear corn, because at this stage it is practically free from ear worms, which prefer more tender plant food. On the exterior outer husk cover there is seldom any blackening to indicate interior infection. The husks next to the interior of the ear are, in severe cases, covered by a black dust of *Aspergillus* spores, which is confined to the layers nearest to the grains. As already stated above, the black mold of ear corn follows injuries made by the corn-ear worm. It should be added that in Texas other insects, as well as rodents and birds, are all responsible for opening the way to black mold infection. The corn weevil, which is also troublesome to ear corn, does not seem to play any rôle in favoring infection, because the weevils attack only mature ears, especially under poor storage conditions. The symptoms on broom corn are decidedly different from those on the ear corn. The heads of broom corn when affected by the yellow and the black molds have a dirty black-and-yellow appearance, the grains becoming covered with fungus threads of both *Aspergillus niger* and *A. flavus*, and frequently also by numerous other organisms, especially *Fusarium*, which give the broom corn heads a decidedly moldy appearance (Figure 3, c and d) and a characteristic stale odor. Frequently, as a result of the molds, the broom corn becomes shriveled and lose their germinating power very early. As with molds of ear corn, the molds of broom corn are favored by the attack of insects or by improper curing. Furthermore, as already mentioned, some varieties are more susceptible than others.

CAUSE OF THE BLACK AND THE YELLOW MOLDS.

The fact that *Aspergillus niger* and *A. flavus* are always associated with black and yellow molds of ear corn and broom corn in Texas would indicate that these organisms are the cause of the trouble. From a scientific consideration more proof is necessary. It is extremely easy to isolate both *Aspergillus niger* and *A. flavus*, since both of these fungi fruit abundantly on their hosts. All that is necessary is to take a bit of tissue from the affected host and drop it in sterilized water. Dilution plates from this water, which contains the spores of these two organisms, will yield pure cultures of *A. niger*, *A. flavus*, or both. From the plate cultures, the two organisms may readily be transferred pure to slanted tubes, and then used for inoculation purposes.

In order definitely to establish the cause of the black and the yellow molds of ear corn, artificial inoculations were begun on ears both in the field on the growing plants, and on cut ear corn brought into the laboratory. Inoculations were made on ears in every stage of development; that is, those which were barely formed to those which were

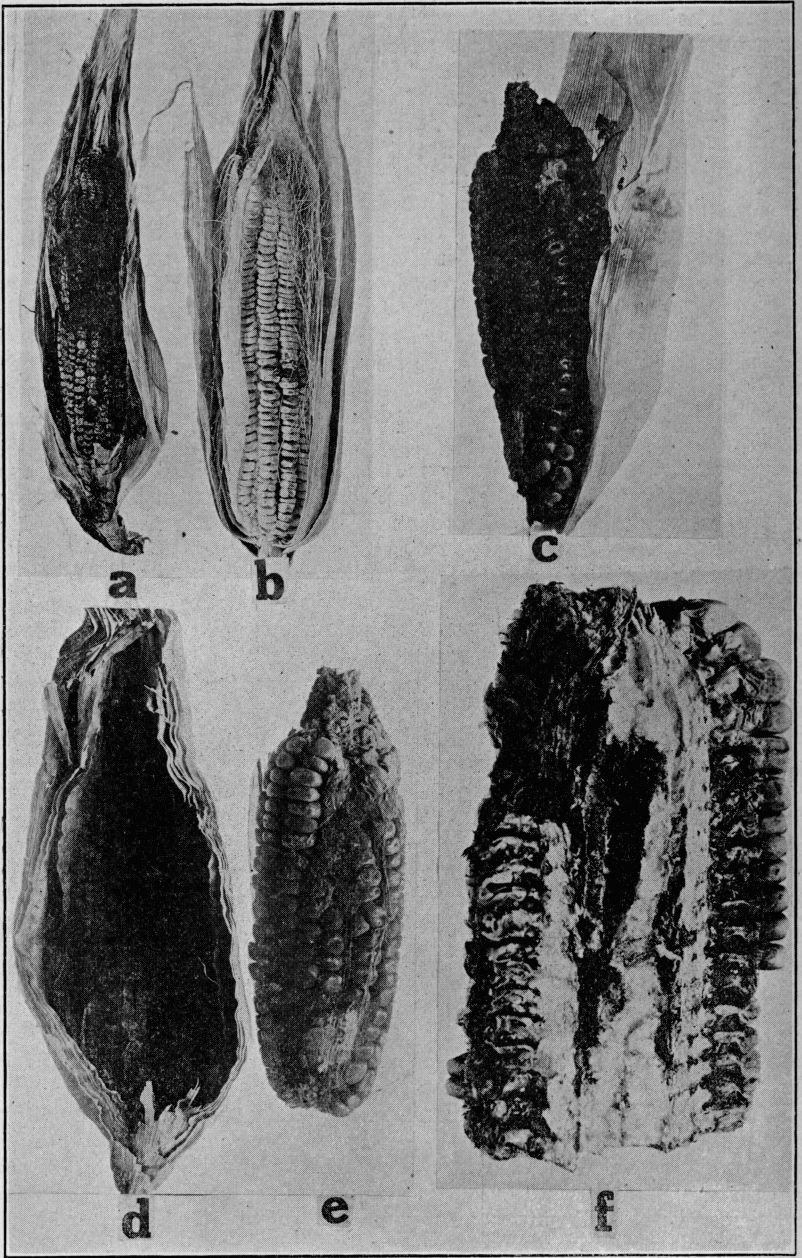


FIGURE 2.

a. and *b.* Two young ears of corn attacked early by the ear worm and later destroyed by black mold. The grains failed to fill out properly. *c.* Ear corn totally destroyed by black mold. *d.* Shucks of ear corn in which the cob has been removed to show the blackening of the husks by the black mold. *e.* Ear corn destroyed by black mold same as *d.* from which it has been removed. *f.* Young ear corn cut longitudinally to show black mold working into the cob.

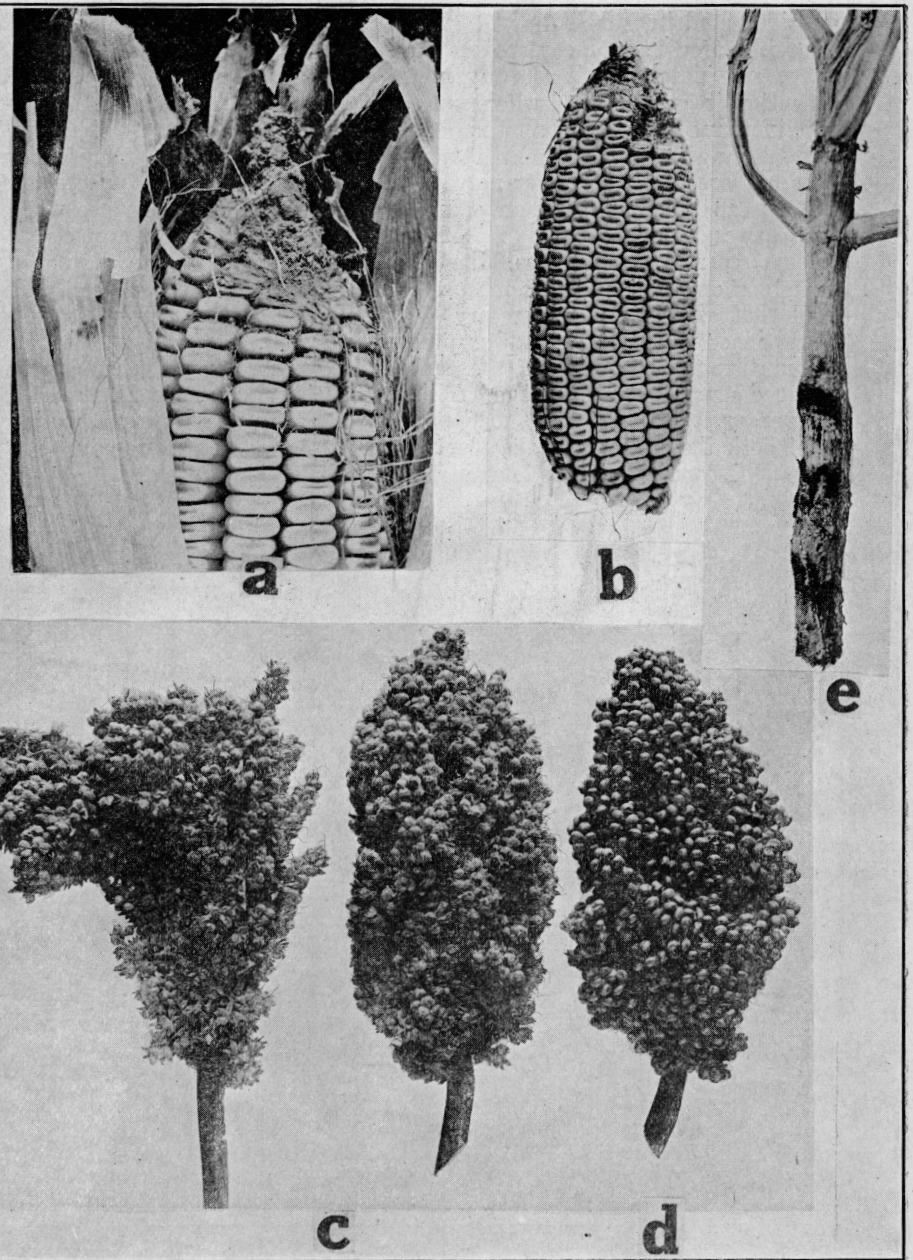


FIGURE 3.

a and b. Ear corn affected by yellow mold, which nearly always works at the tip of the ear. e. Cowpea affected by *Aspergillus niger* at the base of the stem. c. Two heads of broom corn affected by black mold. d. Healthy head.

fully matured. The methods of inoculation and the results obtained are indicated in Tables 1 to 5. From these tables, it is clearly evident that infection can take place on ear corn only when it is in its milky stage and not when it is fully matured and hardened. Furthermore, infection is only possible when spores of *Aspergillus niger* and *A. flavus* are introduced through a puncture in the soft tender ear. Occasionally *Aspergillus* spores may start to grow at the place of infection where there is no needle puncture, but here, however, the fungus does not spread internally and only develops superficially at the place where the spores are deposited. No inoculations were tried on any of the broom corn. Nevertheless, the results obtained on ear corn definitely show that ripe and mature corn is not subject to the ear mold and that an injury is necessary for infection. This is what actually takes place in the field, and, as previously stated, the injury is brought about through the work of the ear-worm or other insects when attacking the ear in its milky stage. This is especially emphasized because of numerous statements made to the writer by various growers who believe that both the yellow and the black molds of ear corn are brought about when the corn is left too long in the field after harvesting.

Table 1.—Field inoculation of ear corn, milky stage.

No. of Ears Inoculated	Method of Inoculation	Date of Inoculation	Source of Aspergillus	Result of Inoculation after 20 Days
7	By puncture through husks and injuring grains*	June 3, 1918†	<i>A. niger</i> from corn ears.....	All 7 ears thoroughly infected.
12	By puncture through husks and injuring grains*	June 3, 1918†	<i>A. niger</i> from cotton bolls.....	All 12 ears thoroughly infected.
19	By puncture through husks and injuring grains*	June 3, 1918†	<i>A. niger</i> from pomegranates..	5 ears thoroughly infected, remainder only lightly infected.
16	By puncture through husks and injuring grains*	June 3, 1918†	<i>A. niger</i> from peanut.....	All 16 ears thoroughly infected.
10	By puncture through husks and injuring grains*	June 3, 1918†	<i>A. niger</i> from broom corn.....	All 10 ears thoroughly infected.
8	By puncture through husks and injuring grains*	June 3, 1918†	<i>A. flavus</i> from corn.....	4 ears medium infected, remainder lightly infected.
7	By puncture through husks and injuring grains* No spores used.	June 3, 1918†	Check.....	All healthy.

*Inoculated ears enclosed in inverted thick paper bag and clasped by rubber band. This was done to create moist surroundings.

†Ears inoculated on corn plants in the author's home garden at College Station.

Table 2.—Field inoculation of ear corn fully ripened.

No. of Ears Inoculated	Method of Inoculation	Date of Inoculation	Source of Aspergillus	Results of Inoculation After One Month
5	By puncture through husks and injuring grains*	Aug. 20, 1918†	<i>A. niger</i> from ear corn.....	No infection.
3	By puncture through husks and injuring grains*	Aug. 20, 1918†	<i>A. niger</i> from broom corn.....	No infection.
10	By puncture through husks and injuring grains*	Aug. 20, 1918†	<i>A. niger</i> from onion.....	No infection.
5	By puncture through husks and injuring grains* No spore used.	Aug. 20, 1918†	Check.....	No infection.
3	By puncture through husks and injuring grains*	Aug. 20, 1918†	<i>A. flavus</i> from ear corn.....	No infection.
12	By puncture through husks and injuring grains*	Aug. 20, 1918†	<i>A. niger</i> from broom corn.....	No infection.

*Inoculated ears enclosed in inverted thick paper bag.

†Inoculation carried out in corn field at Prairie View College, Prairie View, Texas.

Table 3.—Detached Ear Corn, milky stage.†

No. of Ears Inoculated	Method of Inoculation	Date of Inoculation	Source of <i>Aspergillus</i> Used	Result of Inoculation After Twenty Days
62	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from ear corn	60 ears thoroughly infected.
44	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from onion	44 ears thoroughly infected.
22	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from squash fruit	19 ears thoroughly infected.
27	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from broom corn heads	27 ears thoroughly infected.
17	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from cotton bolls	15 ears thoroughly infected.
25	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from peanut	21 ears thoroughly infected.
28	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from pomegranate	20 ears thoroughly infected.
15	By puncture through husks and injuring grains*	June 3, 1918	<i>A. niger</i> from cowpea	10 ears thoroughly infected.
26	By puncture through husks and injuring grains*	June 3, 1918	<i>A. flavus</i> from corn	21 ears infected, infection localized to punctured area.
19	By puncture through husks and injuring grains*	June 3, 1918	<i>A. flavus</i> from broom corn heads	19 ears infected, infection localized to punctured area.
10	By puncture through husks and injuring grains*	June 3, 1918	<i>A. flavus</i> from dead bee larvae	10 ears infected, infection localized to punctured area.
12	Spores applied on grains under husks and without scratch or puncture.*	June 6, 1918	<i>A. niger</i> from corn ear	5 showing slight infection.
18	Spores applied on grains under husks without injury but with water introduced.	June 6, 1918	<i>A. niger</i> from corn ear	All ears thoroughly infected
27	Spores applied through puncture in husks but without injury to grain.*	June 7, 1920	<i>A. niger</i> from corn ear	19 mildly infected.
22	Spores applied through puncture in husks but without injury to grain and without placing in moist chamber.	June 7, 1918	<i>A. niger</i> from corn ear	16 mildly infected.

*Kept in moist chamber for three days.

†Ear corn from Prairie View Normal.

Table 4.—Detached ear corn, semi-ripe.

No. of Ears Inoculated	Method of Inoculation	Date of Inoculation	Source of <i>Aspergillus</i> Used	Result of Inoculation After Twenty Days
18	By puncture through husks and injuring grains*	June 8, 1918	<i>A. niger</i> from ear corn	Weak infection on all 18 ears at place of inoculation.†
25	By puncture through husks and injuring grains*	June 8, 1918	<i>A. niger</i> from broom corn	Weak infection on 21 ears at place of inoculation.
16	By puncture through husks and injuring grains*	June 8, 1918	<i>A. niger</i> from peanut	Weak infection on all 16 ears at place of inoculation.
21	By puncture through husks and injuring grains*	June 10, 1918	<i>A. niger</i> from pomegranate	Weak infection on 17 ears at place of inoculation.
10	By puncture through husks and injuring grains*	June 10, 1918	<i>A. flavus</i> from ear corn	Very weak infection on all 10 ears.
17	By puncture through husks without injury to grain.*	June 8, 1918	<i>A. niger</i> from ear corn	Very weak infection on 4 ears.
11	By puncture through husks and injuring grains	June 12, 1918	<i>A. niger</i> from ear corn	Very weak infection on 3 ears.
9	By puncture through husks and injuring grains	June 12, 1918	<i>A. flavus</i> from broom corn	No. infection.
16	Spores applied on grains under husks without puncture or scratch.	June 13, 1918	<i>A. niger</i> from ear corn	Infection extremely weak at point of inoculation, not spreading.
12	Spores applied on grains under husks without injury, 5 drops of sterile water applied at point of inoculation.	June 13, 1918	<i>A. niger</i> from ear corn	Very weak infection at point of inoculation, fungus not spreading.

*Kept in moist chamber for three days.

†Infection localized to interior of husk as indicated by a black powder at an area of about one-half to one inch diameter at place of inoculation. Infection was more restricted on the corn grains immediately under the husk.

Table 5.—Detached ear corn fully ripened.

No. of Ears Inoculated	Method of Inoculation	Date of Inoculation	Source of Aspergillus Used	Results of Inoculation After Eight Weeks.
16	By puncture through husks and injuring grains*	Sept. 5, 1918	<i>A. niger</i> from corn ear.....	Trace of Aspergillus growth at place of inoculation.
24	By puncture through husks and injuring grains*	Sept. 5, 1918	<i>A. niger</i> from peanut.....	Trace of Aspergillus growth at place of inoculation.
14	By puncture through husks and injuring grains*	Sept. 5, 1918	<i>A. niger</i> from corn ear.....	No infection.
24	By puncture through husks and injuring grains*	Sept. 12, 1918	<i>A. niger</i> from corn ear.....	No infection.
26	Liberal dusting spores between husks and grain but no scratching.	Sept. 14, 1918	<i>A. niger</i> from corn ear.....	No infection.
21	Liberal dusting spores between husks and grain but no scratching.	Sept. 20, 1918	<i>A. niger</i> from onion.....	No infection.
10	Husks removed, ear dipped in sterile water containing suspension of spores.*	Sept. 20, 1918	<i>A. niger</i> from ear corn.....	Traces of superficial infection at points of injury.
19	Husks removed, grains scratched and treated as above.*	Sept. 20, 1918	<i>A. niger</i> from ear corn.....	Traces of superficial infection at points of injury.
11	Husks removed, grains scratched and treated as above.*	Sept. 22, 1918	<i>A. niger</i> from ear corn.....	No infection.
12	Placed mouldy ear between healthy ones, husks removed in all and kept dry.	Sept. 26, 1918	<i>A. niger</i> from ear corn.....	No infection.
11	Same as above.....	Sept. 26, 1918	<i>A. niger</i> from ear corn.....	No infection.

*Kept in moist chamber for three days.

The younger the ear during infection, the darker it is when harvested because at this early stage the fungus has had an ideal medium on which to grow. Furthermore, the moisture under the husk of the young ear seems to favor the fruiting of *A. niger* and *A. flavus*, both of which readily shed their spores, giving the characteristic black or yellow dusty appearance to the affected ears. When infection takes place at an early stage of the development of the ear, *Aspergillus niger*, especially, may also gain access to the interior of the cob and give it a black charred appearance. The black color of the cob is brought about primarily by the color of the spores of the fungus, the latter of which sporulates profusely in the interior of the tissue just as readily as it does on the surface of the grains under the husks. The mycelium of both species of *Aspergillus* is white to yellowish. Sporulation seldom takes place on the outer surface of the husk, for which reason it is not always easy to tell the amount of ear-mold infection until after harvesting and shucking.

IS BLACK MOLD A STORAGE DISEASE?

To further determine whether or not both the yellow and the black molds may be spread to healthy mature corn in the crib, the following experiment was carried out during the winters of 1918 and 1919: In each case, 10 bushels of healthy ear corn in the husk were secured from Prairie View, Texas. This corn grew on good land and developed into normal, well-formed ears. The corn was also well matured, as it was allowed to stay in the field until late in the fall and was gathered when there was no rain. These ears, having been husked and stored in a crib, were arranged into layers of (a) healthy, and (b) infected layers. The ears in each layer were properly counted and allowed to remain undisturbed during the entire winter in a crib well protected from rain. Early in the spring, the corn was taken out and carefully examined. It was found that the same number of healthy as well as black moldy ears were taken out as were originally placed in the crib. This proved conclusively that the black mold of ear corn does not spread in the crib even though diseased ears are intermixed with healthy ones, provided the corn is kept dry. Similarly, three bushels of healthy mature corn secured from Prairie View were intermingled with diseased ears affected with the black mold and stored away in a crib. The corn was sprinkled with water once every two weeks, so as to keep it moist, and the ears were left undisturbed during the entire winter. Early in the spring, they were taken out and examined. It was found that numerous species of *Fusaria* and *Aspergillus niger* were growing on the exterior husks giving them a somewhat black, dusty appearance. *Aspergillus*, however, did not penetrate the interior as to injure the ears, which were fairly ruined by *Fusarium*. This indicated that moisture does not seriously encourage black mold on mature corn as it does *Fusaria*. This also agrees with the results of the artificial inoculations as reported in Table 5, and with field observations. In the field, mature corn when exposed to wet-weather conditions may be ruined by *Fusaria*, but not by *Aspergillus*. It is not intended here to give the impression that there is no harm in keeping stored corn under moist conditions. Such a practice is never carried out. The point brought out mainly is that the black mold of corn does not spread in the corn crib under dry storage conditions. This further strengthens the proof that black mold is a disease which follows insect injury in the field, and

infection only takes place when the ears are in a milky stage and not after they are well matured.

CARRIERS OF BLACK MOLD OF EAR CORN

To establish the relationship of the corn-ear worm (*Chloridea obsoleta*) to the black mold of ear corn, the following experiments were tried out: A large number of corn-ear worms were collected from a corn field. These worms were gathered from young tassels and young ears, and brought to the laboratory in sterilized test tubes containing sterilized water. These were then placed on an electric revolving machine and shaken for 20 minutes. The water from these vials was then plated out in ordinary petri plates containing agar agar. Of the many organisms isolated were *Aspergillus niger* and *A. flavus*, *Penicillium*, *Fusarium*, and a large number of bacteria. The *Aspergillus* isolated seemed to be identical with *A. niger* and *A. flavus* originally obtained from diseased ear corn. This proves that the ear worm does carry on the exterior of its body spores of *Aspergillus* as well as of other fungi. A large number of ear worms were also collected from the tomato and cotton bolls and treated in the same way as those collected from ear corn. *Aspergillus* and numerous other organisms were isolated from the water from the worms thus collected and reinoculated in young ear corn, cotton bolls, and tomato fruit (see Table 6).

Aspergillus flavus and more especially *A. niger* are frequently found on cotton bolls which in the field were previously attacked by the cotton boll worm. The latter often causes considerable damage to cotton bolls, and invariably boll rot (*Pseudomonas malvacearum*) follows the injury of the boll worm thus causing the boll to become softened and to decay. At this stage, it is very common to find *Aspergillus niger* invading the diseased bolls, which turn black, on account of the spores of this fungus, which fruits abundantly on the outer surface as well as in the interior of the boll tissue and lint. Frequently, also, *Aspergillus niger* invades the cotton boll immediately after it has been injured by the boll worm, in which case the affected boll becomes semi-dry rotted and later shrivels and blackens. With tomatoes, when the fruit is injured by boll worm, *Aspergillus niger*, although frequently found at the place of injury, does not seem to cause a decay as long as the tomatoes are green and in a growing condition. As shown in Table 6, however, tomato fruit, when detached from the vines, will rot if inoculated in the laboratory with spores of *Aspergillus niger* or *A. flavus*. In transit, when the tomatoes are packed poorly or carelessly, or when fruit infested with the ear worm is shipped they will often black-rot as a result of the secondary invasion of *Aspergillus niger*. Hence it is seen, from an economic point of view, that *A. niger* and *A. flavus* are important not only as causing the black and the yellow molds of ear corn and broom corn, but also as being a semi-parasite on cotton bolls, tomato fruit and other plants.

Table 6.—Aspergillus inoculation of hosts other than ear corn.†

Source of Organism	Host Inoculated	Result of Inoculation	Symptoms Produced
<i>A. niger</i> from peanut	Picked bean pods	30% infection, slow rotting	Soft rot, tissue firm.
<i>A. niger</i> from cotton boll	Picked bean pods	18% infection, slow rotting	Soft rot, tissue firm.
<i>A. niger</i> from peach	Picked bean pods	28% infection, slow rotting	Soft rot, tissue firm.
<i>A. niger</i> from onion	Picked bean pods	26% infection, slow rotting	Soft rot, tissue firm.
<i>A. niger</i> from corn ear	Picked bean pods	21% infection, slow rotting	Soft rot, tissue firm.
<i>A. niger</i> from corn ear	Growing bean plant 12 months old*	Negative, all healthy	
<i>A. niger</i> from onion	Growing bean plant 12 months old*	Negative, all healthy	
<i>A. flavus</i> from ear corn	Growing bean plant 12 months old*	Negative, all healthy	
25 checks	Picked bean pods	All healthy	
<i>A. flavus</i> from corn ear	Picked bean pods	26% infection, slow rotting	Soft rot.
<i>A. flavus</i> from broom corn	Picked bean pods	16% infection, slow rotting	Soft rot.
<i>A. flavus</i> from dead bees	Picked bean pods	24% infection, slow rotting	Soft rot.
<i>A. flavus</i> from peanut	25 tomato fruit half ripe	79% infection, rapid rotting	Leak, soft rot.
<i>A. flavus</i> from cotton boll	25 tomato fruit half ripe	67% infection, rapid rotting	Leak, soft rot.
<i>A. flavus</i> from peach	25 tomato fruit half ripe	83% infection, rapid rotting	Leak, soft rot.
<i>A. flavus</i> from onion	25 tomato fruit half ripe	71% infection, rapid rotting	Leak, soft rot.
<i>A. flavus</i> from corn ear	25 tomato fruit half ripe	64% infection, rapid rotting	Leak, soft rot.
25 checks	25 tomato fruit half ripe	Negative, all healthy	None.
<i>A. niger</i> from corn ear	6 tomato plants 10 weeks old*	Negative, all healthy	None.
<i>A. niger</i> from onion	6 tomato plants 10 weeks old*	Negative, all healthy	None.
<i>A. flavus</i> from corn ear	6 tomato plants 10 weeks old*	Negative, all healthy	None.
<i>A. flavus</i> from broom corn	6 tomato plants 10 weeks old*	Negative, all healthy	None.
<i>A. flavus</i> from broom corn	Cucumber fruit, half ripe	Negative, all healthy	None.
<i>A. flavus</i> from corn ear	Cucumber fruit, half ripe	Negative, all healthy	None.
<i>A. niger</i> from corn ear	Cucumber fruit, half ripe	Negative, all healthy	None.
<i>A. niger</i> from onion	Cucumber fruit, half ripe	Negative, all healthy	None.
<i>A. flavus</i> from corn ear	Cucumber fruit, half ripe	Negative, all healthy	None.
<i>A. flavus</i> from broom corn	Cucumber fruit, half ripe	Negative, all healthy	None.
<i>A. flavus</i> from broom corn	7 cucumber plants 5 weeks old*	Negative, all healthy	None.
<i>A. flavus</i> from corn ear	7 cucumber plants 5 weeks old*	Negative, all healthy	None.
<i>A. niger</i> from corn ear	7 cucumber plants 5 weeks old*	Negative, all healthy	None.
<i>A. niger</i> from onion	7 cucumber plants 5 weeks old*	Negative, all healthy	None.
<i>A. niger</i> from corn ear	20 squash fruit, full grown	50% infection	Soft rot. Rotting slow.
<i>A. niger</i> from onion	20 squash fruit, full grown	50% infection	Soft rot. Rotting slow.
<i>A. flavus</i> from corn ear	20 squash fruit, full grown	15% infected	Soft rot.
<i>A. flavus</i> from broom corn	20 squash fruit, full grown	18% infected	Soft rot.
<i>A. flavus</i> from broom corn	8 squash plants, 6 weeks old*	No infection	None.
<i>A. flavus</i> from corn ear	8 squash plants, 6 weeks old*	No infection	None.
<i>A. niger</i> from onion	8 squash plants, 6 weeks old*	No infection	None.
<i>A. niger</i> from corn ear	30 carrots, fully grown	10% infection, slow rotting	Solid, water soaked rot.
<i>A. niger</i> from onion	30 carrots, fully grown	14% infection, slow rotting	Solid, water soaked rot.
<i>A. niger</i> from peanut	30 carrots, fully grown	19% infection, slow rotting	Solid, water soaked rot.
<i>A. flavus</i> from corn ear	30 carrots, fully grown	6% infection, slow rotting	Solid, water soaked rot.
<i>A. flavus</i> from broom corn	30 carrots, fully grown	12% infection, slow rotting	Solid, water soaked rot.

<i>A. flavus</i> from broom corn.....	25 onions, fully grown bulbs.....	Negative, no infection.....	None.
<i>A. flavus</i> from corn ear.....	25 onions, fully grown bulbs.....	Negative, no infection.....	None.
<i>A. niger</i> from onion bulb.....	25 onions, fully grown bulbs.....	68% infection, medium slow rot.....	Semi-soft rot of outer scales of bulb.
<i>A. niger</i> from corn ear.....	25 onions, fully grown bulbs.....	73% infection, medium slow rot.....	Semi-soft rot of outer scales of bulb.
<i>A. niger</i> from peanut.....	25 onions, fully grown bulbs.....	59% infection, medium slow rotting.....	Semi-soft rot of outer scales of bulb
<i>A. niger</i> from dead bees.....	25 onions, fully grown bulbs.....	69% infection, medium slow rotting.....	Semi-soft rot of outer scales of bulb
<i>A. niger</i> from cotton boll.....	25 onions, fully grown bulbs.....	48% infection, medium slow rotting.....	Semi-soft rot of outer scales of bulb
<i>A. niger</i> from pomegranate.....	25 onions, fully grown bulbs.....	64% infection, slow medium rotting.....	Semi-soft rot of outer scales of bulb
<i>A. niger</i> from pomegranate.....	25 tubers Irish potatoes, var. Mountain green.	28% infection, slow rotting.....	Moist hard rot, infected tissue.
<i>A. niger</i> from ear corn.....	25 tubers Irish potatoes, var. Mountain green.	34% infection, slow rotting.....	Moist hard rot, infected tissue.
<i>A. niger</i> from cotton.....	25 tubers Irish potatoes, var. Mountain green.	30% infection, slow rotting.....	Moist hard rot, infected tissue.
<i>A. niger</i> from onion.....	25 tubers Irish potatoes, var. Mountain green.	30% infection, slow rotting.....	Moist hard rot, infected tissue.
<i>A. niger</i> from peanut.....	25 tubers Irish potatoes, var. Mountain green.	22% infection, slow rotting.....	Moist hard rot, infected tissue.
<i>A. flavus</i> from corn ear.....	25 tubers Irish potatoes, var. Mountain green.	19% infection, slow rotting.....	Moist hard rot, infected tissue.
<i>A. flavus</i> from broom corn.....	25 tubers Irish potatoes, var. Mountain green.	23% infection, slow rotting.....	Moist hard rot, infected tissue.
25 checks.....	25 tubers Irish potatoes, var. Mountain green.	Negative, all healthy.....	None.
<i>A. niger</i> from onion.....	50 picked pea pods.....	14% infection, slow rotting.....	Semi-soft rot.
<i>A. niger</i> from ear corn.....	50 picked pea pods.....	10% infection, slow rotting.....	Semi-soft rot.
<i>A. flavus</i> from ear corn.....	50 picked pea pods.....	12% infection, slow rot.....	Semi-soft rot.
<i>A. flavus</i> from ear corn.....	10 pea plants 8 weeks old.....	Negative, no infection.....	None.
<i>A. niger</i> from ear corn.....	10 pea plants 8 weeks old.....	Negative, no infection.....	None.

†Inoculated July 16, 1918; final observation made after 20 days.

*Kept in moist chambers.

AIR AND DUST CARRIERS OF ASPERGILLUS SPORES.

From the foregoing discussion, it is evident that the boll worm or corn-ear worm and possibly other insects not experimented with, not only open the way to *Aspergillus* infection, but that they are also carriers of the spores of these fungi. These worms undoubtedly gather the spores from the dust on the plant with which their bodies come in contact. That dust does contain spores of *Aspergillus* was proved by the following experiment: Immediately after a wind storm, dust was carefully gathered, with a sterilized camel's hair brush, into a sterilized test tube, from various foliage of truck plants in the garden, more especially from corn, tomatoes and beans. Some of this dust was then plated out in the usual way and, among the numerous organisms which appeared in the plate cultures, at least one-fifth of the growth was *Aspergillus*. Furthermore, the air itself carries with it among many other organisms, spores of *Aspergillus niger* as well as *A. flavus*. To determine this, 50 poured petri plates were made and after properly cooling and solidifying, were taken out in the open and the lids removed for one-half minute. After a period of 24 hours incubation, these plates showed a varied mycological flora, among which was also *Aspergillus niger* as well as *A. flavus*. From this it is easy to realize that as soon as worms cause an injury to the corn ears or to any other plant material which they attack, a way is opened to possible secondary infection from *Aspergillus* and other fungi. With the yellow mold, *A. flavus*, as already stated, the fungus seems to attack only the tips of ears of those varieties which grow upright. In this case it is very easy for dew or rain water to lodge in the husk, and any spores of *Aspergillus flavus* which may have been introduced with the dust will find an ideal medium to start growth.

SUSCEPTIBLE HOSTS.

As already referred to on page 3, and from a study of Table 6, it will be seen that besides the corn and broom corn, both *Aspergillus niger* and *A. flavus* are capable of infecting a large number of hosts. Mention has already been made (5, 18, 19, 20, 24, 33, 37, and 38) that *Aspergillus niger* is responsible for diseases on various plants. In Texas the pomegranate is seriously attacked by *Aspergillus*, but this fruit is not grown to any extent to make it of sufficient economic importance. Nevertheless, the plant is grown for ornamental purposes on a scale large enough to warrant consideration. During dry seasons, and as soon as the pomegranates become partly mature, a large percentage of them split open and immediately rot from *Aspergillus* (Figure 4, a and b). The splitting of the fruit is common during dry weather. The *Aspergillus* fungus, no doubt, enters the fruit through mechanical or insect injury. As regards the onion, *Aspergillus niger* is of considerable economic importance in Texas. This is true not so much as a field trouble but more so in storage or in transit. The disease is especially serious on the large Spanish type of Denia onions. The black mold first appears as blackened areas on the upper two or three layers of the bulb scales (Figure 4, e). When carefully examining these dark areas, one will find that they consist of deadened tissue, the surface of which has been blackened with loose spores of *Aspergillus niger*. As the diseased area increases, larger numbers of the bulb scales become

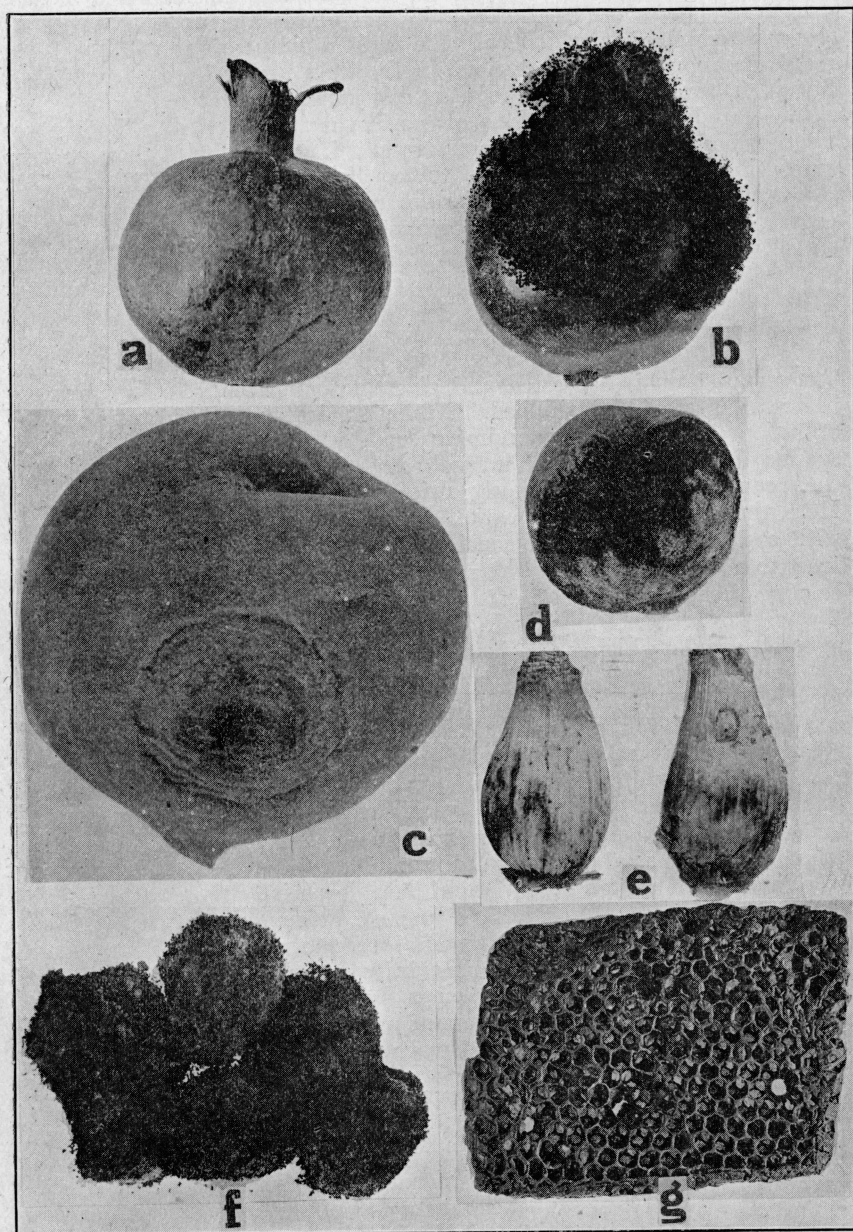


FIGURE 4.

a. Healthy pomegranate. *b.* Pomegranate affected by black mold (natural infection).
c. Peach artificially inoculated with *Aspergillus niger*. *d.* and *f.* Peach fruit destroyed by
A. niger in transit. *e.* Young onion bulbs spotted by *A. niger*. *g.* Young bee colony the larvae
of which were apparently killed by *A. niger* and *A. flavus*.

involved and in time the entire onion shrivels and blackens and is then worthless. Frequently, also, a soft rot follows the opening made by *Aspergillus niger* in which case the bulb rapidly soft-rots. The black mold of onions is seldom serious in the field under Laredo conditions, but it is severe when the crop is dug during a wet spell and when shipped under poor conditions of ventilation. Onions dug during wet weather become gorged with moisture and it is such bulbs that are very susceptible to the black mold. According to Wayne (37 and 38), *Aspergillus niger* in Ohio attacks not only the mature onions, but also causes considerable damage to both seeds and sets. This, as already stated, is not the case in Texas and it is probably due to our peculiar soil and climatic conditions, which are different from those of Ohio.

On the peanut, black mold does not seem to be of any importance as a field disease in Texas. It often, however, causes serious loss in storage or in transit. With this crop, black mold is to be looked for when the peanut crop is harvested and given no opportunity to dry properly before it is stored or shipped. Numerous instances have come to the attention of the author wherein car loads of shipped peanuts have been spoiled during transit and in such cases it has been determined that the peanuts were not properly dried prior to loading and shipping. Here the exterior of the peanut shells becomes blackened and coated with a dark powder which consists of the spores of *Aspergillus niger*. In extreme cases, the causal fungus also invades the inside of the nuts, causing them to blacken and to shrivel thus becoming unfit for seed or for oil purposes. In general, however, oil mills refuse altogether to accept peanuts which show infection on the exterior of the hull. Although of no considerable economic importance, *Aspergillus niger* is frequently found to injure Irish potatoes in Texas, causing a semi-dry rot. Here the fungus seems able to penetrate the tuber only through a cut or bruise and it frequently becomes serious in transit or under poor storage conditions. In referring to Table 6, one will see that under laboratory conditions. *Aspergillus* is able to cause a rot on detached bean and pea pods, as well as on apples, peaches, (Figure 4, c, d and f), plums, and carrots (Figure 5, h). In the field, it is not uncommon to find pea pods (Figure 5, c) especially those of black-eyed peas and bean pods (Figure 5, d and e), thoroughly rotted by *Aspergillus niger*. In this case, however, the fungus is not able to attack sound pods but only follows insect injury, especially that of the boll worm, which, in Texas, feeds on a large number of hosts. As to peaches, the author does not know of a single case wherein *Aspergillus niger* was found to cause a rot on peaches in the orchard, although it commonly follows brown rot, *Sclerotinia fructigena*, in transit. Nevertheless, as seen from Table 6 and Figure 4, c, d, and f, *Aspergillus niger* will readily grow on green as well as on mature detached peaches and cause them to rot. Infected peaches are at first water-soaked and then shrink and become blackened. Frequently the fruit melts away, owing no doubt, to the invasion of secondary organisms. The same is true of apples, which become rotted although the tissue remains more or less firm and the fungus seems to fruit more in the interior of the tissue than on the outside. This does not seem the case with the affected peaches as the fungus fruits abundantly on the outer surface of the fruit only. From a consideration of the trucking industry, *A. niger* is economically important in the sense that it produces a serious rot of squashes and pumpkins in the field. Affected squashes become soft-

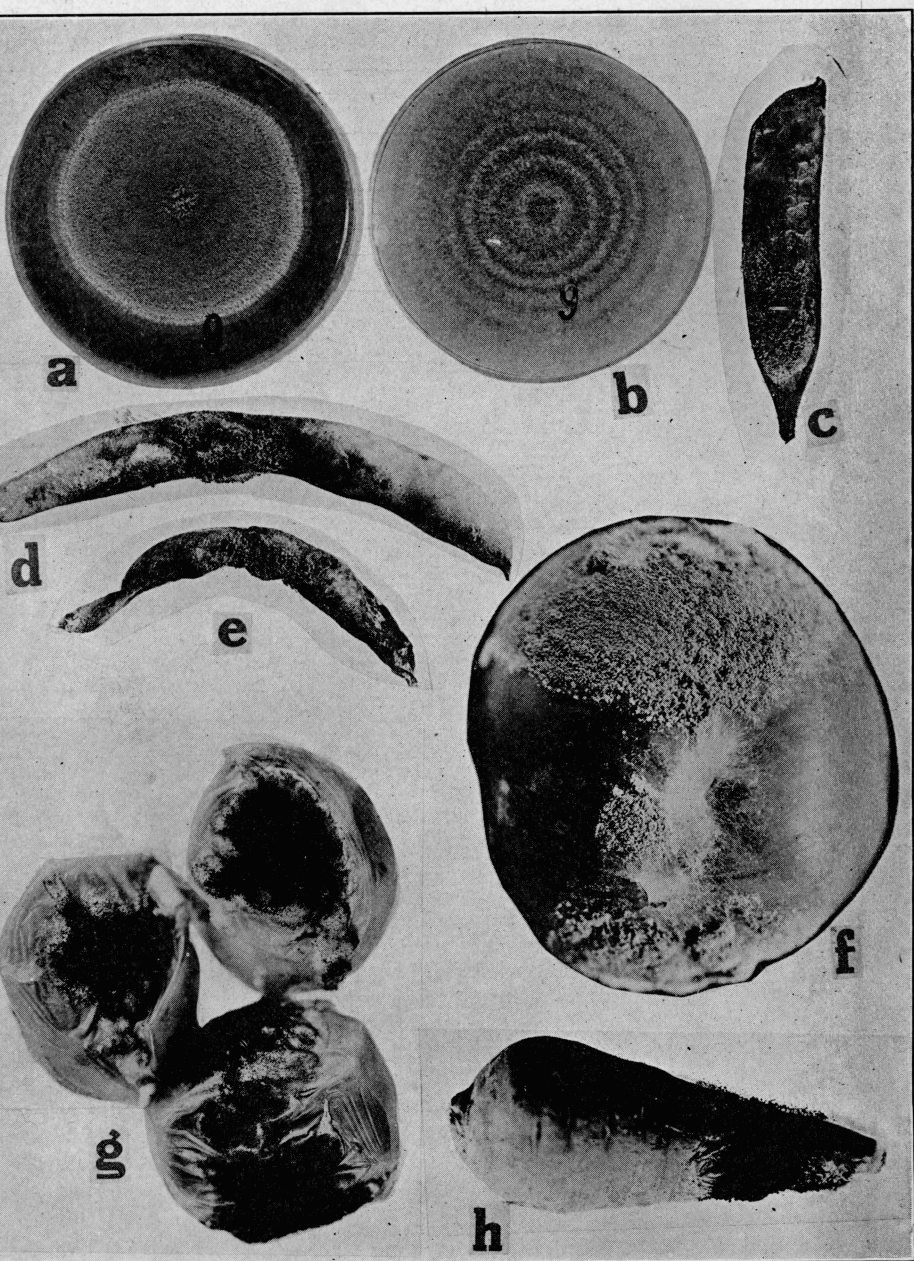


FIGURE 5.

A. flavus on hard potato agar. *b.* *A. flavus* on hard bean agar. *c.* Garden pea pod artificially inoculated by *A. niger*. *d.* Bean pod artificially inoculated with *A. niger*. *e.* Bean pod artificially inoculated *A. flavus*. *f.* Tomato fruit artificially inoculated by *A. niger*. *g.* Carrot artificially inoculated by *A. niger*.

rotted, water-soaked, and in a day or two are black, owing to the appearance of the fruiting heads of the fungus. Infection, no doubt, follows insect injury or starts on the dead corolla where *A. niger* begins as a saprophyte and winds up as a parasite.

Not only does the *Aspergillus* group contain species which are harmful to plants, but there are also those which are capable of producing serious diseases in farm animals. According to Mayo (23) Enzootic cerebritis of horses in Kansas is attributed to *Aspergillus glaucus*. This is likewise supported by Dalrymple (9) and by Schoenleber (32), who claim that cerebritis of horses is probably the result of feeding moldy corn which is affected with *Aspergillus glaucus*. Haslam (17) claims that blind staggers (Meningoencephalitis) in Kansas is caused by feeding moldy corn. Bayer (1) cites a case of cattle poisoning due to feeding of moldy clover invaded by *Aspergillus glaucus*. Pearson and Ravenel (27) report a case of Pneumomycosis due to *Aspergillus fumigatus*. Mohler and Buckley (26) report a case of pulmonary mycosis on birds attributed to *Aspergillus fumigatus*. They also claim that this same fungus would cause a disease in chickens, pigeons, rabbits, and guinea pigs. Likewise, Savouré (30) attributes a serious tubercular lesion (internal mycosis) in animals, to *Aspergillus fumigatus*, *A. oryzae*, and *A. sulphureus*. Likewise, Bodin and Savouré (4) claim to have tested out *Aspergillus fumigatus*, *Aspergillus niger* and *Sterigmatocystis pseudonigra* on guinea pigs and found that *A. fumigatus* alone was pathogenic. Moreover, Sartory and Jourde (29) also found *Aspergillus fumigatus* and *Sterigmatocystis* spp. to be pathogenic to rabbits.

It is interesting to note in this connection that the pathogenicity of some species of *Aspergillus* may be utilized in the control of insect pests. Valerio and Jongh (10 and 11) have tested out the effect of *Aspergillus niger* and *Aspergillus glaucus* on the control of mosquito larvae, *Culex* and *Anopheles*. They found that species of *Aspergillus* were pathogenic to mosquito larvae, but that in practice it did not pay to use these organisms on a large scale. Garrett (13) found an *Aspergillus* species to be pathogenic on the sugar cane mealy bug (*Pseudococcus calceolariae*) in Louisiana. Harshberger (16) states that *Aspergillus flavus* is capable of causing a disease on silk worm cocoons. In Texas, the author has repeatedly isolated *Aspergillus niger* and *Aspergillus flavus* from dead bee larvae obtained from diseased bee hives (Figure 4, g). The pathogenicity of these two organisms on bees, however, was not tried out. Gilruth (14) reports a red discoloration of butter due to *Aspergillus nidulans*. Likewise, Butjagin (6) reports the spoiling of meat due to *Aspergillus niger*.

From the above discussion of the literature, it is seen that there are strong evidences that *Aspergillus glaucus* is the cause of serious complications in animals and in but one instance is it claimed specifically that *A. niger* induces a disease on mosquito larvae. Inasmuch as the black mold of ear corn and broom corn in Texas is caused by *Aspergillus niger*, it is not certain whether such corn, when fed to horses, would bring about a disease similar to that produced by *Aspergillus glaucus*, as is reported by Mayo (23) and others to occur in Kansas. This being a veterinary question, no attempt was made to determine the effect of feeding black moldy corn to horses. The author, however, may state definitely that he has fed corn infected with black mold to adult chickens for more than two months without any apparent harmful

effect to their health. The general belief among farmers is that when moldy corn is fed to horses, it will give them the heaves. Others claim that it will be the cause of serious coughs. The author takes no responsibility for these statements. Reed and Barber (28) express the belief that *Aspergillus fumigatus* is not responsible for stock poisoning when this fungus occurs on moldy silage.

PHYSIOLOGICAL SPECIES

It has already been stated that the black mold of corn is caused by a fungus, *Aspergillus niger*, which can gain entrance to the ear only through a puncture and especially when the ear is in its milky stage. In the field, the injury which favors infection is that caused by the corn-ear worm and other agencies mentioned on pages 15 and 18. It has been further stated that *Aspergillus niger* also causes a mold on peanuts, onions, and other crops. The author has repeatedly isolated *Aspergillus niger* not only from ear corn, but also from peanuts, cotton bolls, cowpea pods, onions, pomegranates, Irish potato tubers, squashes and broom corn. It became necessary, therefore, to determine whether or not strains of the black *Aspergillus* isolated from the various hosts mentioned above are distinct or physiological species, or whether they are one and the same as *Aspergillus niger*, which causes the black mold of corn. Accordingly, strains isolated from the above hosts were inoculated in ear corn, the method of inoculation being indicated in Table 6, which shows that strains of *Aspergillus niger* isolated from peanuts, onions, pomegranates, cotton bolls, cowpeas, Irish potatoes, squashes and broom corn will produce a black mold on ear corn when inoculated in its milky stage as readily as will the strain of *Aspergillus* isolated from the ear corn itself. This seems to prove that there does not exist any physiological species or strains in the fungus known as *Aspergillus niger*.

CONDITIONS FAVORING INFECTION.

It has been made clear that ear mold of corn is caused by the fungus *Aspergillus niger* and that this organism can only invade the ear during its milky stage and when it has been injured by the ear worm. Since black mold in the field depends primarily on the injury of the ear worm or other insects, it follows therefore that the severity of the black mold will be correlated with the severity of the presence of the ear worm. Garman and Jewett (12) claim that the ear worm is not as common in some seasons as it is in others. It may, in fact, be prevalent in one locality one season and yet be practically rare another season. This seems to be the condition in Texas, where the ear worm is more prevalent during dry seasons. During 1917 and 1918, two dry summers, the ear worm was unusually severe and as a result, the injury from the black mold of corn was in the same proportion. On the other hand, during 1919, which was practically a wet season, there was very little ear worm and consequently correspondingly very little black mold. Bishop (3), on the other hand, states that throughout the entire country 70 to 90 per cent. of the ears of field corn are attacked (referring to the ear-worm). Following this injury, molds frequently gain access to the ears and damage them still further. This is especially true during wet seasons. Bishop does not state which molds he has in mind. It is, of course, conceivable that during wet weather, various molds should

attack corn ears which were previously injured by the ear worm or by other insects. As far as the black mold of corn is concerned, dry-weather conditions do not prevent infection of *Aspergillus niger*, since the latter directly follows the injury made by the ear worm. It seems plain that the greatest amount of black mold of corn should be looked for during dry seasons. Moreover, during wet weather, the corn-ear worm, although prevalent, is parasitized by numerous other insects and fungi which tend to keep it in check. This also indirectly results in reduced losses from black mold. As to the yellow mold, as already stated, infection depends directly on the amount of moisture which is able to collect at the tip end of the ear as it stands erect on the stalk.

Table 7.—Effect of healthy and moldy corn seed.

Source of Seed	Number of Trials	Number of Seed per Pot	Per cent of Germination	Effect of Growth
From healthy ears.....	10	50	100	Normal.
Infected ear, grains apparently normal but cob blackened and entirely invaded by <i>Aspergillus niger</i>	17	50	48	Seedlings weak eventually becoming stunted and dying.
Badly infected ear, grain small, shriveled and under-sized....	48	50	2 to 12	Seedlings weak, dying at an early age.
From healthy ear, healthy ground cob worked in soil.....	12	50	100	Decided stimulation over healthy check.
From healthy ear, quantity of spores of <i>A. niger</i> worked in soil.	8	50	100	Seedlings normal at first becoming stunted, yellowing and dying.

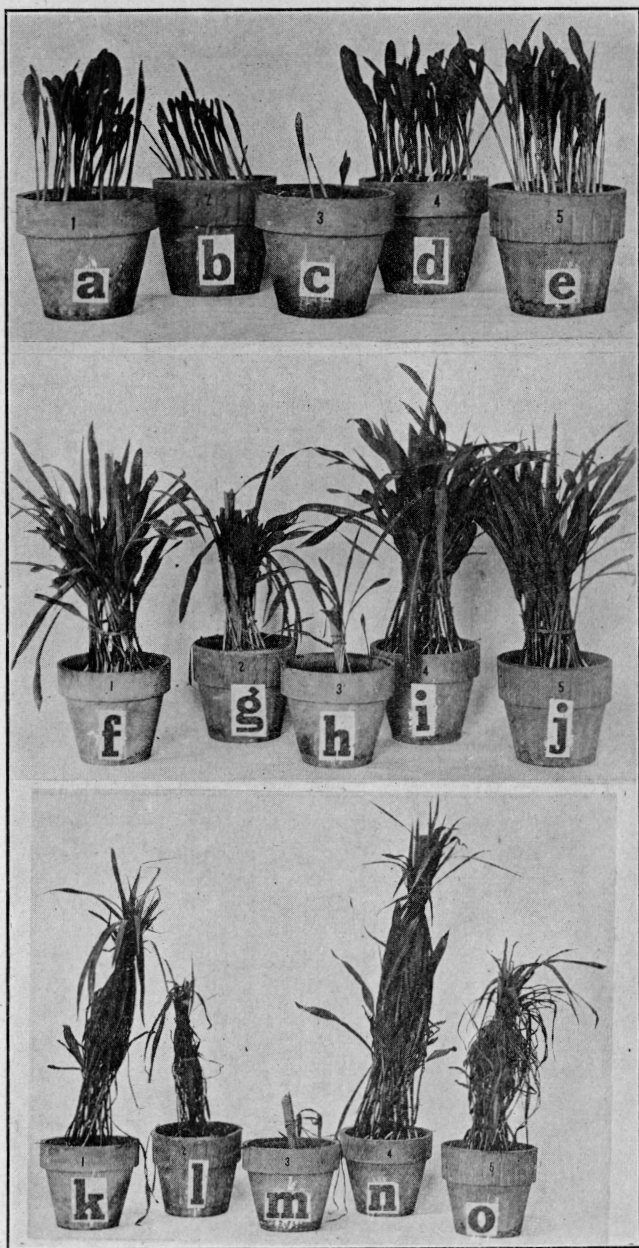


FIGURE 6.

a. Check pot planted with healthy corn seed. b. Pot planted with corn seed taken from an apparently healthy ear but with cob affected by *A. niger*. c. Pot planted with seed from a badly blackened ear in which both cob and seed were invaded by *A. niger*. d. Pot planted with healthy seed in which crushed up cob from a healthy ear was worked in. e. Pot planted with healthy corn seed in which crushed cob from a diseased ear was worked in. f, g, h, i and j. The same as a, b, c, d and e except that the plants are one week older. Notice the weak seedlings in pot g, and the same in pot h. There seems also a slight stimulation in pot i, and a checking of growth in pot j. k, l, m, n, and o. the same as f, g, h, i, and j, except that the plants are two weeks older. Notice the healthy check plants in pot k and decided stimulation of growth in pot n, and the dying of the plants in pots l, m, and o.

USE OF BLACK MOLDY SEED CORN.

It has been frequently asked whether or not it is safe to plant corn affected with black mold. Experiments have been carried out to determine whether or not such a practice is desirable or safe. It is taken for granted that no one will ever use seed corn that came from badly diseased and shriveled ears. Nevertheless, there are cases of late infection, where the ears were but slightly injured by the black mold, infection being confined to the place of injury from the ear worm and resulting in an ear more or less normally developed and apparently containing well nourished and fully developed grains, although their surface may be covered with spores of *Aspergillus niger*. The question might, therefore, be asked whether such grains are safe for planting purposes. To determine this the following experiments were planned: Seed from moldy corn was secured from various sources and arranged as follows: (1) seeds from healthy ears; (2) seed corn apparently normal, but taken from an ear whose cob was thoroughly blackened and invaded by *Aspergillus niger*; (3) seed corn taken from ears which were badly affected and shriveled as a result of black mold. The seeds from these three sources were each planted separately in pots in order to determine their germination. In connection with this, healthy seed corn was planted in pots into which was worked healthy ground cob. Finally more healthy seed corn was secured and planted in pots in soil into which was worked a large amount of spores of *Aspergillus niger* secured from diseased ear corn. The results of this experiment are shown in Table 7. In studying Table 7 and in referring to Figure 6, a to e, one will see that check pot Figure 6 a was normal in germination. In pot b, germination was normal, but the seedlings were weak. In pot c, the germination was poor and there was a very weak stand; pot d, normal germination and apparent slight stimulation; pot e, normal germination and apparent slight stimulation in some of the seedlings. In further reference to Figure 6, f to j, it is seen that as the seedlings progressed in age, they lagged in pot g, while those in pot h were decidedly dwarfed. The seedlings in pot i were apparently the strongest of all, owing, it seems, to the addition of the ground healthy corn cob. Whether the cob in this case acted as a fertilizer or not has not been determined. In further reference to Figure, 6 k to o, it is seen that the check in pot k made considerable headway and the plants in pot n were still leading. On the other hand, the seedlings in pots l and m were practically dead, whereas seedlings in pot o were partly dead. This clearly demonstrates that for seed purposes, only grains from healthy ears should be secured. Furthermore, no matter how sound a grain appears, it should never be taken for seed if it came from an ear which had the least infection or if the cob has been decayed from black mold. Furthermore, at no time should seed corn be taken from badly diseased ears, as in this case not only will the germination be very inferior, but the few seed that will germinate will produce weak plants which will eventually die. Finally, when spores of *Aspergillus niger* or black powder residue from diseased black moldy corn, is added to the soil, the grain corn although healthy when planted will result in weak seedlings which eventually die from rotting of the roots by *Aspergillus niger*. This was the case when the roots of the plants from pot o were examined. It was found that most of them were destroyed and blackened and, upon culturing, proved to be *Aspergillus*

niger. As a conclusion, it must therefore be definitely stated that for seed purposes, only healthy seed of absolutely healthy ears should be secured.

SYSTEMATIC RELATIONSHIP.

In dealing with so variable a group of fungi as *Aspergillus* it is necessary to define the generic relationship of the two species, *Aspergillus niger* and *A. flavus*, with which the present bulletin is concerned. In studying over the literature on *Aspergillus*, we find that Van Tieghem (39) has accepted the name of *Aspergillus niger* for the black species of *Aspergillus*. That *Aspergillus* was confused with other fungi, especially with *Ascophora*, was due mainly to a misconception of some French mycologists who preceded Van Tieghem. Fortunately, however, the studies of Robin and others on *Aspergillus* species which cause ear diseases in humans, have led up to the possibility of distinctly separating the black species of *Aspergillus* from the black *Mucors*. Cramer (7) named some forms of *Aspergillus*, *Sterigmatocystis*. The author agrees with Thom (36) that there is nothing to justify the existence of the separate generic name of *Sterigmatocystis* and that all species of that genus should be referred to as *Aspergillus*. In our present work, all species of *Sterigmatocystis* are referred to as *Aspergillus*. [According to Thom's classification of the black *Aspergillus* group, which the writer accepts as correct, *Aspergillus niger* is a form with both primary and secondary sterigmata, the primary sterigmata being 20-30 μ in length.] In referring to page 18 we will see that McMurrin (24) as well as Hodgson (18 to 20) mention a pomegranate rot due to *Sterigmatocystis castanea*. The *Aspergillus* fungus which attacks pomegranates in Texas does not seem to differ from *Aspergillus niger*, which we have repeatedly isolated from ear mold of corn and both of which readily cross from one host to the other. Hence, as already stated, the genus *Sterigmatocystis* is untenable and the fungus which causes the rot of pomegranates in Texas is referred to in this work as *Aspergillus niger*. The same is also true for the strains of black *Aspergillus* isolated from cotton bolls, peanut, squash, black-eyed pea pods, Irish potato tubers, and dead bee larvae. All of the species are morphologically similar. As to the yellow *Aspergillus*, namely, *A. flavus* Link, it is believed that its generic and specific relationship requires no modification.

CULTURE WORK.

All the strains of *Aspergillus* mentioned in the previous discussion were at first grown on a variety of media. The writer has, however, finally decided on two media, namely, hard potato agar and hard bean agar*. Numerous strains were tried out as to their morphology and physiology.

In referring to Figure 7, a to g, we will see that three-day-old plate cultures of *Aspergillus niger* from squash, onion, corn, peanuts, and cotton, on hard potato agar, showed practically no morphological difference in growth. Young colonies of these strains, as is seen from

*By hard agar is here meant the addition of 30 grams of agar agar to each 1,000 cc. of media.

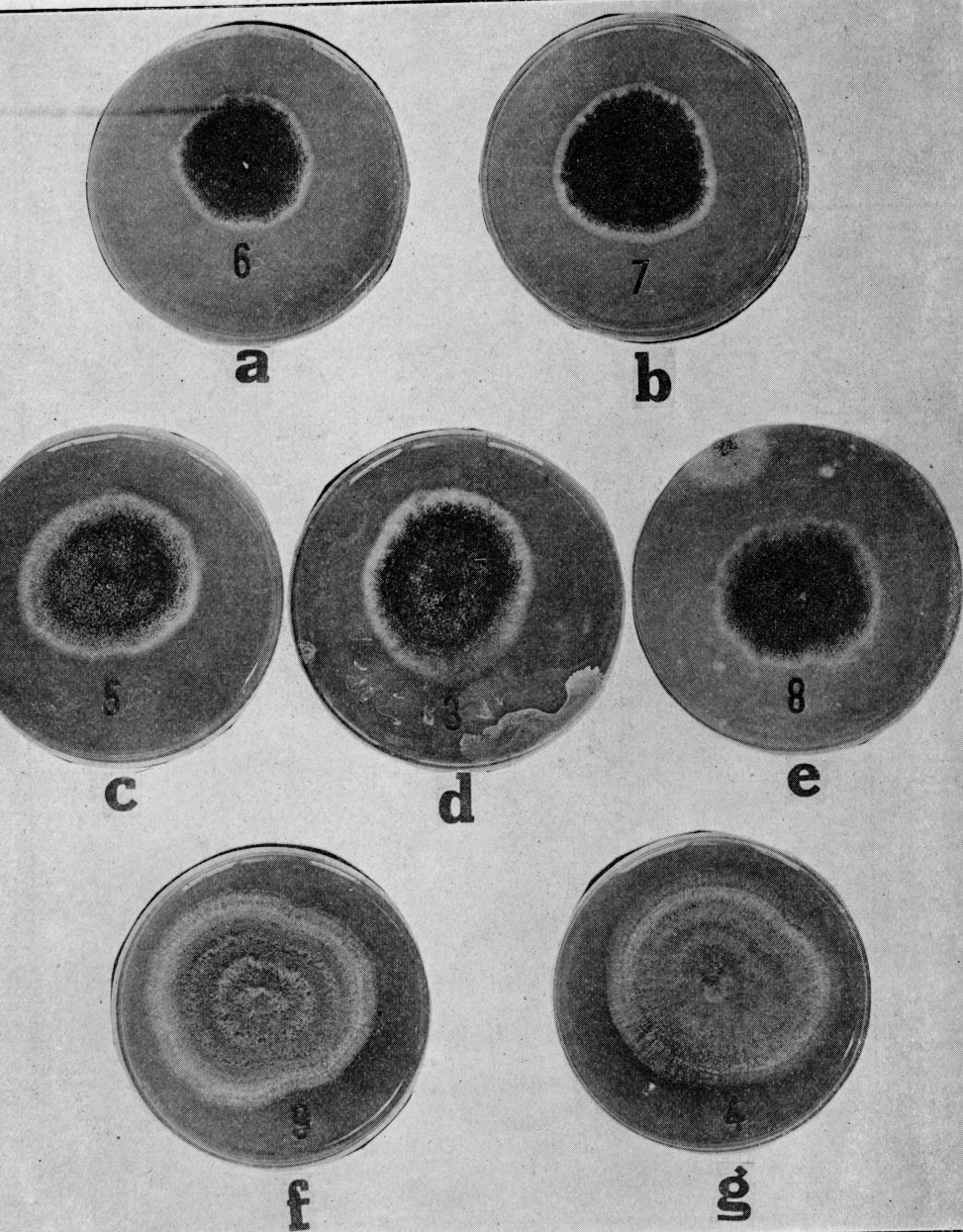


FIGURE 7.

Young plate cultures on hard potato agar of: a. *Aspergillus niger* from cotton bolls. b. *A. niger* from onion bulbs. c. *A. niger* from ear corn. d. *A. niger* from squash fruit. e. *A. niger* from peanut fruit. f. *A. flavus* from ear corn. g. *A. flavus* from dead bee larvae.

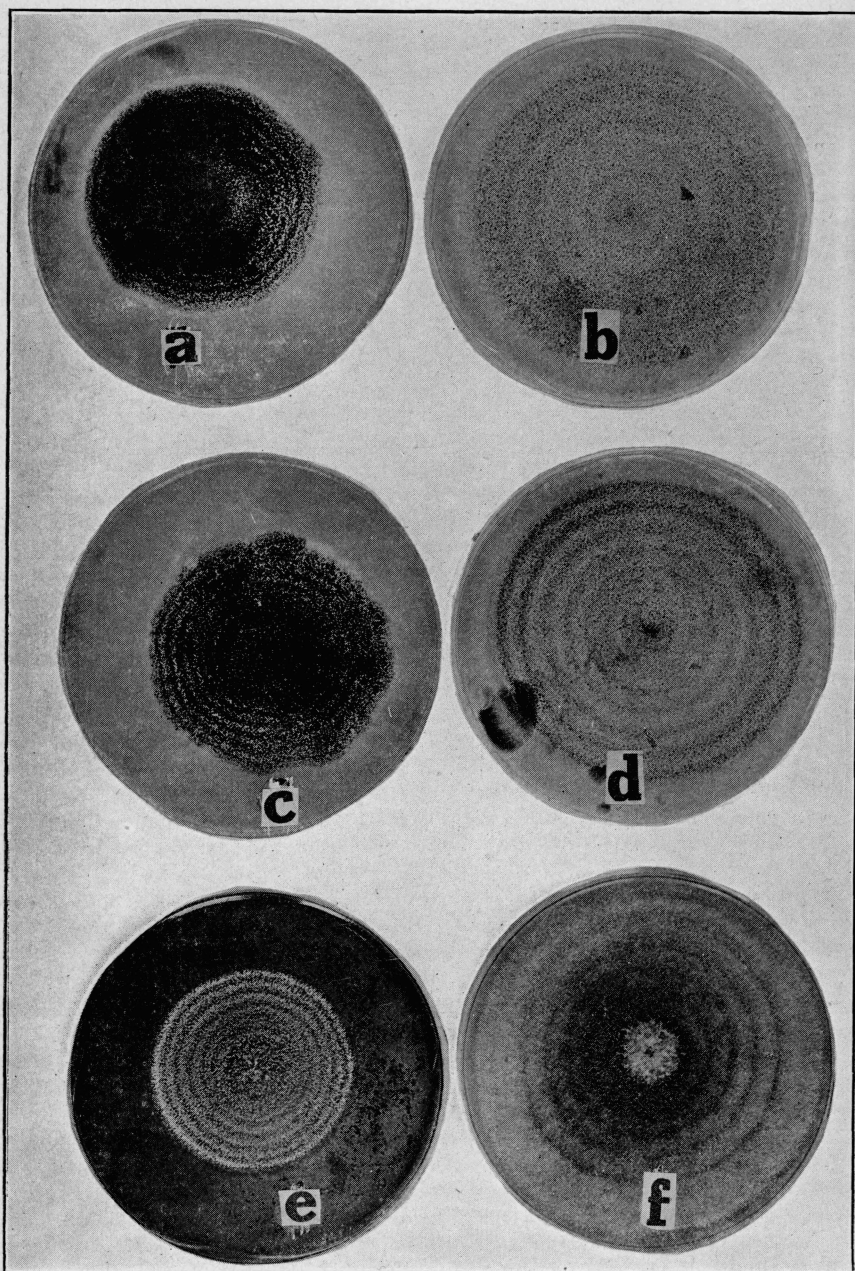


FIGURE 8.

a. and *b.* Advanced plate cultures of *Aspergillus niger* from onion bulb. To the left, on hard potato agar. To the right, on hard bean agar. *c.* and *d.* Advanced plate cultures of *A. niger* from cotton bolls. To the left, on hard potato agar. To the right, on hard bean agar. *e.* and *f.* *A. flavus* from broom corn. To the left, on hard potato agar. To the right, on hard bean agar.

Figure 7, a to e, are compactly zonate with dense numbers of fruiting heads. Likewise, as is seen from Figure 7, f and g, young colonies of *Aspergillus flavus*, isolated from yellow mold of corn, and from dead bee larvae, alike, on hard potato agar are distinctly zonate and the growth not as dense as is the case with *Aspergillus niger*. As the colonies of *A. niger* increase in age, growth becomes more distinctly zonate with a formation of an abundance of spore heads on potato agar. On bean agar, growth is more luxuriant and more distinctly zonate, but with smaller numbers of fruiting heads in the zones (see Figure 8, a to f). Finally, as the same plate cultures become ten to fifteen days old, the outer edge of the colonies on potato agar ceases apparently to zonate, growth becoming irregular, and spore-production apparently ceases or becomes scarce (Figure 9, a to f). This, however, is not the case with the same older strains when grown on bean agar and as seen in Figure 8 (compare a, c, and e to l, d and f). Likewise, older cultures of *Aspergillus flavus* become more intensely zonate on the potato agar but less so on bean agar as seen in Figure 5, a and b. During the course of our culture work, it was noticed that whenever two different colonies of *Aspergillus niger* and *A. flavus* grew near each other, a distinct inhibition line became apparent, preventing the two colonies from meeting and leaving a distinct line of demarcation between the marginal ends (Figure 10, a to e). This line persisted for a long time, often as long as 10 to 15 weeks, but finally the borders of the colonies would gradually intermix. Similar observations were made by Zeller (41) who found the same tendency of colonies of *A. niger* to repel each other.

OTHER DISEASES OF EAR CORN AND BROOM CORN

To clearly distinguish between the black and the yellow molds of ear corn considered in the present bulletin, it is necessary to briefly mention the other diseases with which these two molds are frequently confused.

a. Corn Smut. This is a true smut and is caused by a fungus *Ustilago Zeae* (Beck.) Ung. Corn smut attacks all parts of the plant above ground, namely, leaves, stalks, tassels, and ears. It is recognized as soft boils of various sizes especially on the tassels and ears, in which case the male elements, or the grains in the ears, are replaced by large soft bluish-colored swellings. These when ripe crack open and liberate a black dust which constitutes the spores of the causal fungus. Corn smut cannot be controlled by any method of seed treatment because the fungus is carried over in the soil from year to year and infection does not take place when the corn seed germinate in the soil, but occurs when the plant is in a growing condition and the tissue more or less soft and tender. As already stated, when the smut boils break open, the black, dusty powder is shed on the ear giving it a black, smutty appearance. It is this stage which is likely to be confused with the black mold of ear corn,* the symptoms of which are given on page 5.

Of the diseases which attack broom corn and which may be mistaken for the black mold should be mentioned head smut, which is caused by a fungus *Sorosporium reilianum* (Kuhn.) McAlp. This

*For a description of corn diseases other than black mold, see (35).

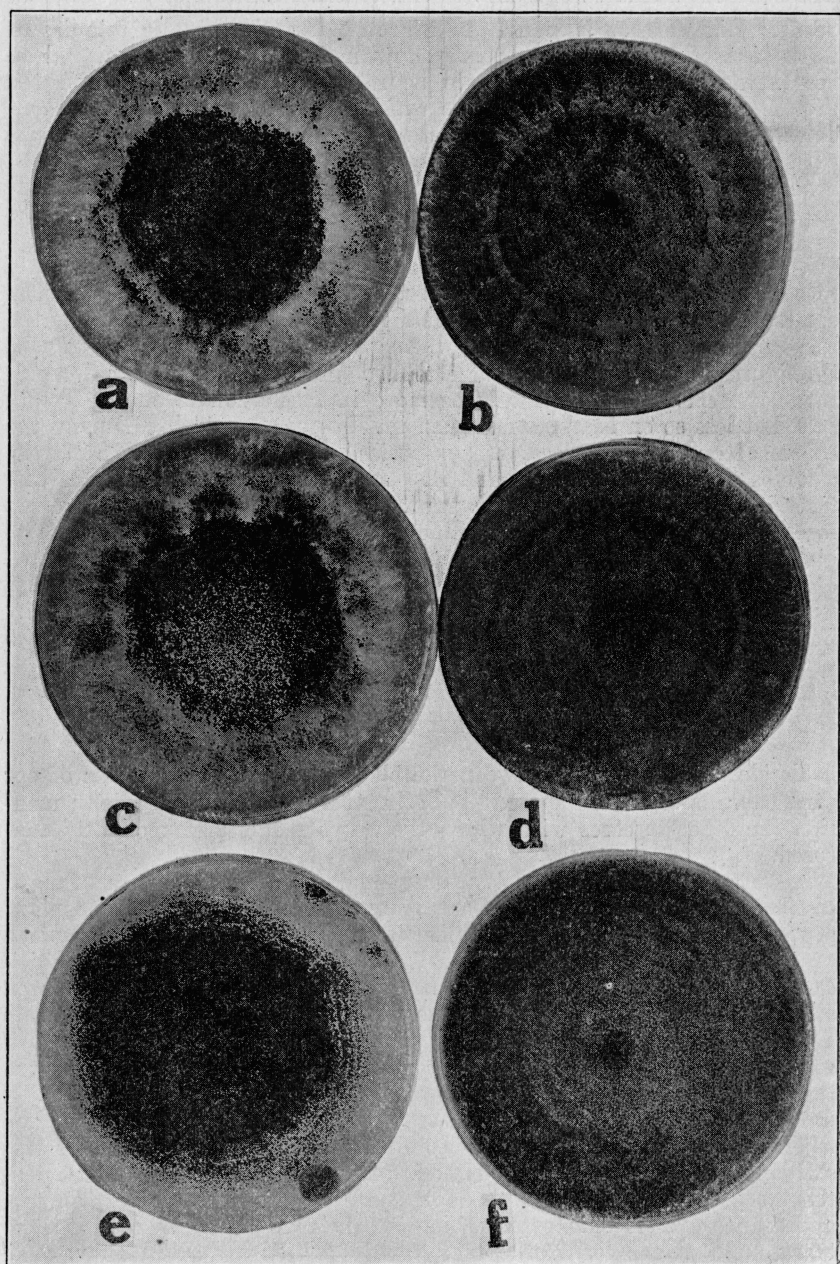


FIGURE 9.

a. and *b.* Old plate cultures of *A. niger* from squash fruit. To the left, on hard potato agar
c. and *d.* Advanced plate cultures of *A. niger* from ear corn. To the left, on hard potato agar
 To the right, on hard bean agar. *e.* and *f.* Advanced plate cultures of *A. niger* from peanut
 fruit. To the left, on hard potato agar. To the right, on hard bean agar.

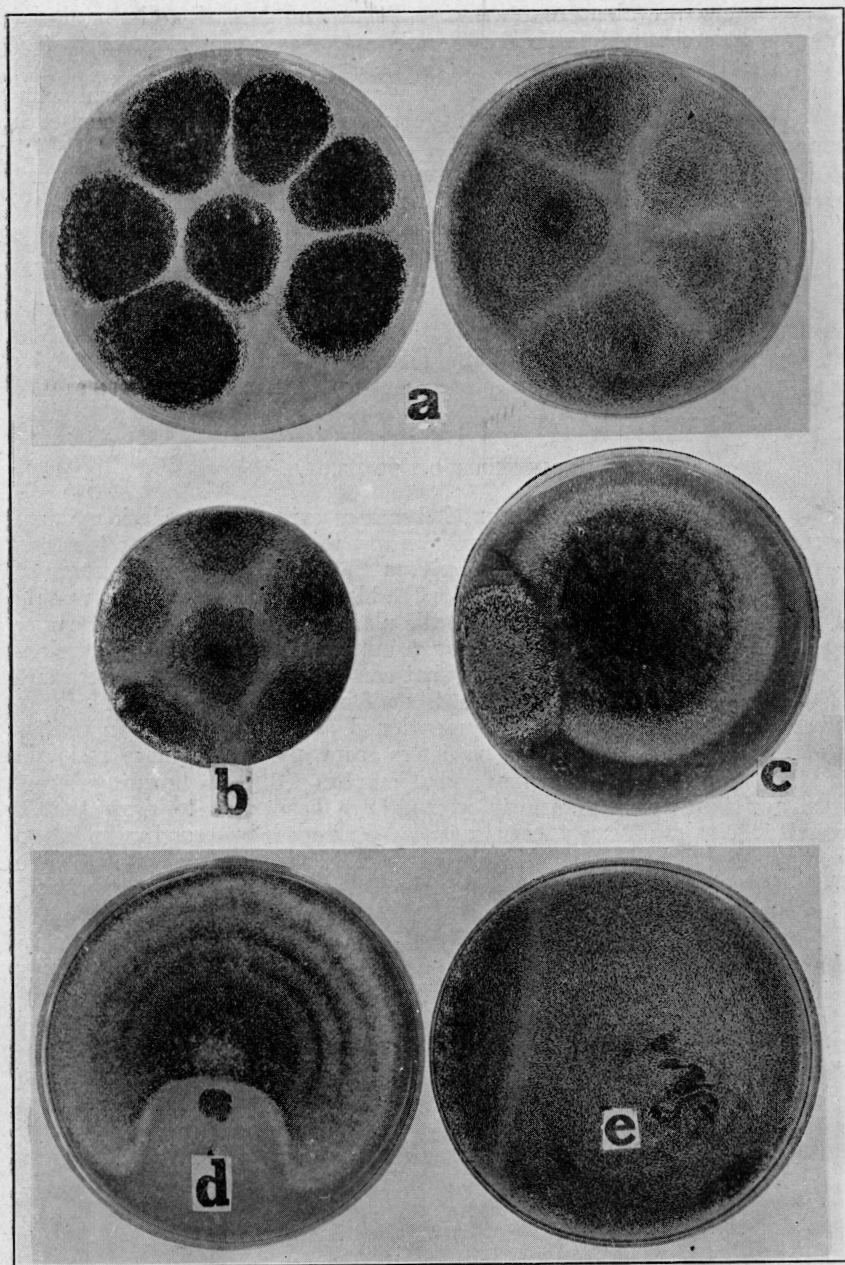


FIGURE 10.

a. Numerous colonies of *A. niger* from ear corn. To the left, on hard potato agar. To the right, on hard bean agar showing a tendency for the colonies to keep away from each other. *b.* Same as *a.* on hard bean agar. *c.* Small colony of *A. flavus* and a large colony of *A. niger* growing in the same petri dish on hard potato agar with the same tendency of the two colonies keeping away from each other. *e.* Same as *a.* *d.* Showing a small yeast colony next to a large colony of *A. flavus*, the former prohibiting the encroachment of the latter.

smut is very prevalent in northwest Texas and is easily distinguished because it is confined to the head which becomes a soft mass of smutty dust; in this respect somewhat resembling true corn smut. Kafir, broom corn, and sorghums are very susceptible to head smut, while the milo seems to be free from it. Like corn smut, head smut of broom corn cannot be controlled by any form of seed treatment.

Another smut which attacks broom corn is that known as kernel smut and is induced by two species of fungi, namely: *Sphacelotheca sorghi* (Lk.) Cl. and *Sphacelotheca cruenta* (Kuhn.) Potter. Kafir, sorgos, broom corn, and Sudan grass are subject to this smut. Infected heads bear a great number of false kernels, which are filled with a mass of black dust consisting of the spores of the causal organism. Kernel smut may be controlled by soaking the seed for two hours in a solution of formaldehyde made up of one pint in 30 gallons of water, or by soaking the seed 10 to 12 minutes in hot water at a temperature of 142 to 143 degrees Fahrenheit.

METHODS OF CONTROL.

From the foregoing discussion, it becomes evident that black mold of corn is dependent directly on insect injury to be able to attack the corn ears. Furthermore, infection is only possible during the milky stage of the ears. Hence control methods for the black mold of ear corn is mainly a problem for the entomologist and the plant breeder; i. e., it is necessary to keep the ear worm in check through cultural methods, and in developing resistant varieties. It seems that some varieties are less attractive to the ear worm and hence are freer from black mold than others. These varieties of corn may be mentioned as Improved Indian Squaw and Surecropper. These two varieties seem also to be more resistant to the yellow mold. Collins and Kempton (8), in breeding sweet corn resistant to the corn-ear worm, have been able to show that a resistant strain of sweet corn may be developed by crossing so as to increase the length and thickness of the husk covering, at the same time reducing the husk leaves. This was accomplished by hybridization and selection of desired plant characters as already stated.

To control yellow mold one has only to plant those varieties of corn that have pendant ears. To do so is especially important in localities with heavy rainfall.

Both the black and the yellow molds on broom corn are also amenable to treatment. Investigations by the Division of Agronomy of the Texas Agricultural Experiment Station have shown that broom corn molds in general, whether black, yellow or white, are more severe on varieties which lack in length of the peduncle. Selections made in this direction have resulted in better broom corn, seeds of which the Division of Agronomy is now ready to distribute to farmers of Texas.

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