

BULLETIN

of the

ILLINOIS NATURAL HISTORY SURVEY

HARLOW B. MILLS, *Chief*

**Fusarium Disease
of Gladiolus:
*Its Causal Agent***

JUNIUS L. FORSBERG



Printed by Authority of the
STATE OF ILLINOIS
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DEPARTMENT OF REGISTRATION AND EDUCATION
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NATURAL
HISTORY SURVEY

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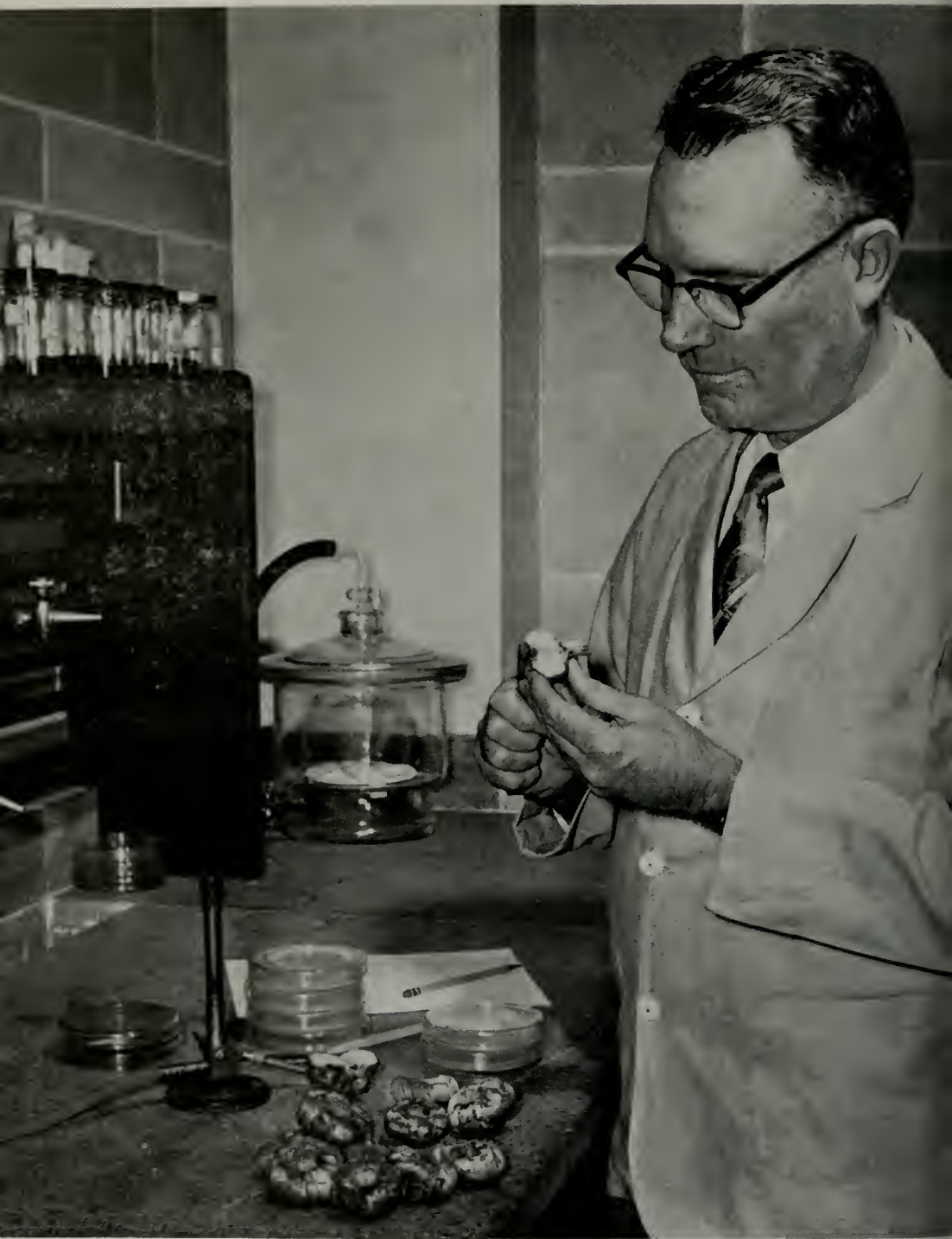
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This paper is a contribution from the Section of Applied Botany and Plant Pathology.

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Examining gladiolus corms for disease symptoms preparatory to making laboratory cultures. The gladiolus *Fusarium* grows well on most of the common laboratory media.

Fusarium Disease of Gladiolus:

Its Causal Agent

JUNIUS L. FORSBERG

THE Fusarium disease of gladiolus is one of the most destructive maladies known to affect flower crops. At a conference held in January, 1953, at Cleveland, Ohio, under the auspices of the Joint Research Committee of the North American Gladiolus Council and the North American Commercial Gladiolus Growers, the concensus of those present was that the Fusarium disease is the most important gladiolus disease in the United States (Ryan 1953). The disease causes large losses to commercial gladiolus growers in Illinois, Indiana, Michigan, California, eastern Washington, and all of Oregon except the northern part. It causes an estimated loss of $1\frac{1}{2}$ to 2 million dollars per year in Florida alone. Because the Fusarium disease is corm-borne and because many of the corms grown in Illinois are shipped to Florida and planted there for winter flower production, the fate of the crop in Florida is of importance to growers in Illinois as well as to growers in Florida. Only one commercial gladiolus-growing area in the United States is not seriously troubled with the Fusarium disease. This is the cool region in western Washington where, according to Gould (1949), Fusarium rot is uncommon except on recently introduced stocks.

ACKNOWLEDGMENTS

The writer wishes to express his sincere appreciation to all who have assisted in any way during the course of this investigation, the greater part of which was presented as a thesis submitted to the Graduate College, University of Illinois, Urbana, in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Plant Pathology. He is especially grateful to the late Dr. Leo R. Tehon, for many years head of the Section of Applied

Botany and Plant Pathology at the Natural History Survey, for his guidance and many helpful suggestions during the course of the investigation and the preparation of the manuscript. Helpful suggestions were made by Dr. Wayne M. Bever, Professor of Plant Pathology, University of Illinois, and Agent, United States Department of Agriculture. The photographic work was done by Mr. William E. Clark, Assistant Technical Photographer of the Natural History Survey, and by Mr. Ray R. Hamm, Manager of the University of Illinois Photographic Laboratory. Several present or former members of the Natural History Survey staff made valuable contributions. Mr. James W. Curfman drew the graphs. Mrs. Rovenia Fitz-Gerald and Mrs. Virginia Lee Johnstone assisted in preparation of the manuscript. Mr. James S. Ayars and Mrs. Blanche P. Young read all or parts of the manuscript. Each offered valuable criticism. The author is especially grateful to his wife, Edith L. Forsberg, for her encouragement and help in many ways during the course of this work.

HISTORY OF THE DISEASE

The disease of gladiolus which is called herein the Fusarium disease is a much misunderstood malady which exists in three forms. These forms have been designated as the vascular, the brown rot, and the basal dry rot types, each of which has been described as a distinct disease by various workers. The disease was first recognized in the early 1920's but it probably existed prior to that time. W. A. Pryal (1909), a California grower, published a note in which he described an interior corm rot and leaf yellowing of gladiolus, but no proof was presented that the disease was caused by a fungus of the genus *Fusarium*.

McCulloch (1944) reported that in 1923 she received from two localities in California a large number of gladiolus corms which, although normal in external appearance, showed, when cut, 90 per cent of the interior rotted. The rot varied from a slight discoloration in the basal scar to browning of the entire core and radiating fibrovascular strands. From these corms was isolated a *Fusarium* that proved capable of causing the disease.

McCulloch (1944) reported further that in 1925 and 1926 she received similar specimens from states as widely separated as North Dakota, Mississippi, and New Jersey. The progress of the disease seemed definitely from west to east, with prevalence increasing each season. In 1926 and 1927, McCulloch found the disease in shipments of corms from Holland. The usual *Fusarium* was isolated from all these specimens.

According to McCulloch (1944), N. van Poeteren "reported the vascular disease as present in Holland as early as 1925." A dry rot of gladiolus caused by *Fusarium* was mentioned in an Annual Report of the Experimental and Research Station, Cheshunt, Hertfordshire, England (Anonymous 1927). Moore (1939) reported a vascular *Fusarium* disease from the same country. Bellard (1933), Dimock (1941, 1945), and Nelson (1937a, 1938a, 1938b, 1948) published brief accounts of the vascular form of the disease. McCulloch (1944) published an extensive account of this form.

Massey (1922) published a brief note and, later (1926), a more extensive description of a corm rot which he considered primarily a disease of stored corms, although infections occurred in the field. McCulloch (1944) considered "the vascular disease" entirely distinct from the corm rot described by Massey.

Nelson (1937b, 1948) described a *Fusarium* disease of gladiolus which he thought distinct from the diseases described by Massey and McCulloch.

McClellan (1947) included as symptom expressions of one disease the symptoms of the two diseases described by Massey and McCulloch. McClellan recognized, however, that there are differences of opinion among those who have worked

with *Fusarium* disease of gladiolus as to whether the two types are distinct diseases or merely forms of the same disease.

NAMES OF THE DISEASE

The use of various names in the literature to designate the forms of the *Fusarium* disease of gladiolus has created much confusion in the minds of readers and research workers. McCulloch (1944) used the names yellows, wilt, and core rot for the vascular type of the disease. Massey (1922, 1926) designated the disease described by him merely as *Fusarium* rot.

Creager (1944) used the name brown rot for the type of corm rot commonly associated with the Picardy variety. He stated, "*Fusarium* brown rot is not the same as *Fusarium* yellows or core rot; they are two distinct diseases." The symptoms commonly found on Picardy, however, seem to be the same as those described by Massey (1926) on other varieties. Picardy was not introduced until 1931, 5 years after Massey's work was published.

Dimock (1945) used only the name yellows and listed Picardy as one of the susceptible varieties.

Nelson (1948) used the names *Fusarium* dry rot and brown rot for the disease originally described by Massey. He stated that this disease "has sometimes been confused with *Fusarium* yellows, an entirely different malady. The chief resemblance between the diseases is that both cause a brown, dry rot of the corm. The best evidence of dissimilarity is demonstrated by the high resistance of the variety Picardy to *Fusarium* yellows and its equally great susceptibility to *Fusarium* dry rot."

The third type of disease described by Nelson (1937b, 1948) was designated by him as basal dry rot.

McClellan (1947) stated, "At least two diseases of gladiolus have been described as being caused by fungi of the *Fusarium* group. One of these is a corm rot that is principally a storage disease; the other is the yellows disease that occurs in the field. Yellows has been subdivided further into (1) a core rot type, a type confined to the water-conducting system; and (2) a basal rot type." He then fol-

lowed with a description of the several kinds of symptoms and considered them as expressions of the same disease.

Magie & Miller (1948) referred to the *Fusarium* corm rot and yellows disease. In a later article the same authors (1949) used the name *Fusarium* brown rot. Magie (1950) used the names *Fusarium* yellows and *Fusarium* corm rot but referred to them as designating a single disease. He

applied the term "yellows symptoms" to the leaf symptoms and vascular discoloration but not to the corm rot phase of the disease.

SYMPTOMATOLOGY

Symptoms of the *Fusarium* disease are produced on foliage, corms, and roots. Detailed descriptions of symptoms associated

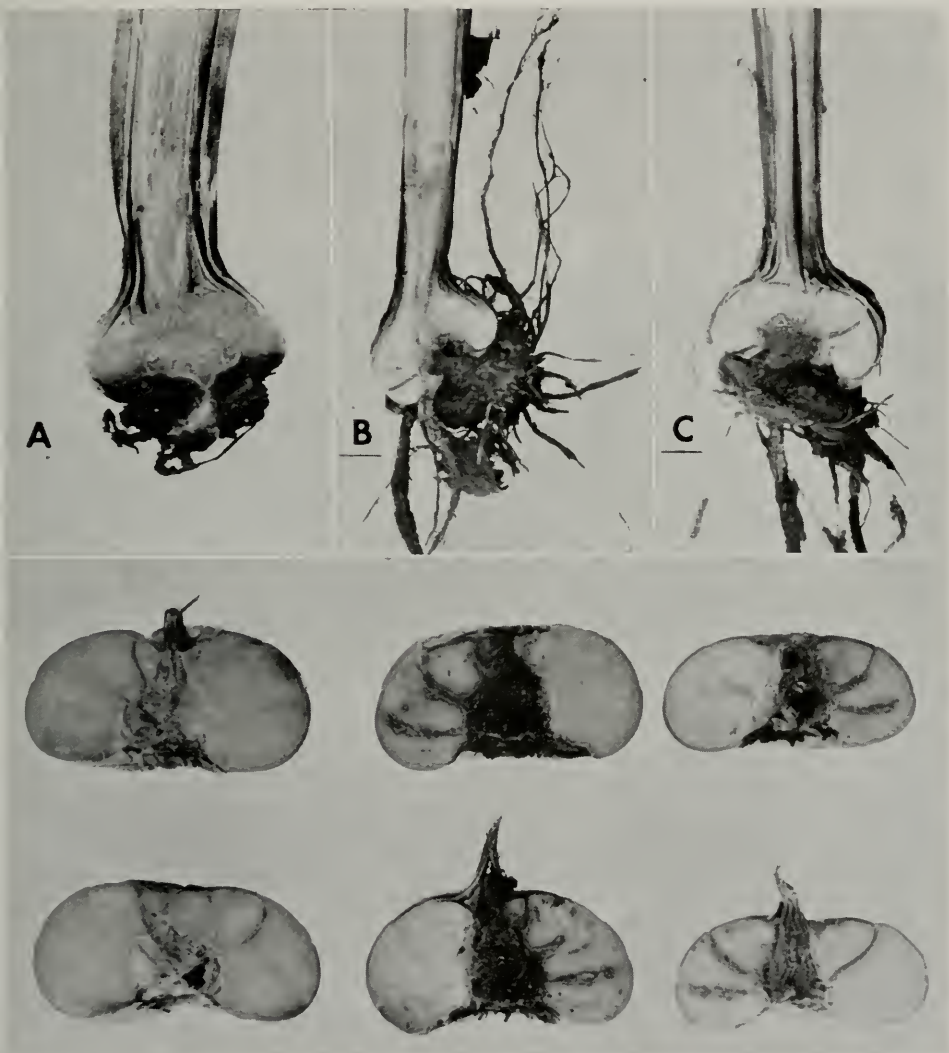


Fig. 1.—Above: lengthwise sections of gladiolus corms showing, *A*, how the brown rot form of the *Fusarium* disease progresses from the mother corm to the daughter corm; *B* and *C*, how the vascular form progresses from the mother corms into the core and vascular tissues of the daughter corms. Below: sections of six older corms showing rotted cores and discolored vascular streaks associated with the vascular form of the disease.

with the three forms of the disease, vascular, brown rot, and basal dry rot, have been published by McCulloch (1944), Massey (1926), Nelson (1948), and McClellan (1947). The symptoms common to all three forms of the disease are a brownish to black dry rot of the corm tissues; yellowing, browning, and death of the foliage; and browning and destruction of the roots.

The three forms of the disease have been distinguished mainly by effects on the corms. In the vascular form of the disease a sectioned corm will reveal a brown discoloration of the core and dark-colored vascular bundles extended laterally into the flesh, fig. 1*B* and *C*. In an advanced stage of the disease, the infected strands reach the surface of the corm at the nodes, and brown lesions develop at these points.

In the brown rot form of the disease, tan, brown, or blackish lesions may occur anywhere on the corm but most commonly near the base, fig. 2. The rotted tissue is often quite thick and may extend all the way through the corm, fig. 2, bottom row. Vascular discoloration is not associated with this form of the disease.

The basal dry rot form of the disease differs from the brown rot form mainly in the thickness and position of the lesions. Basal dry rot lesions occur only on the bases of the corms and are usually restricted to the first and second internodes, fig. 3. The lesions are visible when the corms are dug and, under favorable curing conditions, they do not enlarge after harvest. They rarely, if ever, extend deeper than 2 to 4 millimeters into the flesh. The diseased tissue is dark brown to black,

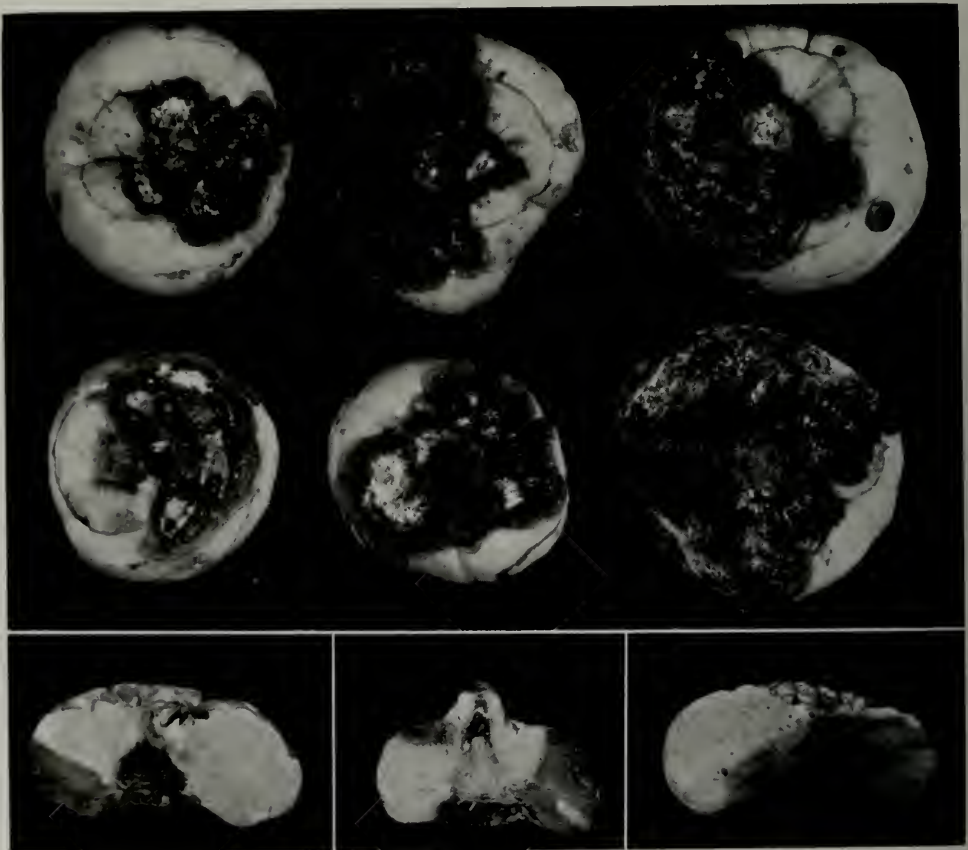


Fig. 2.—Picardy gladiolus corms affected with the brown rot form of the *Fusarium* disease. Above: bottom views of six corms with lesions of various sizes. Below: sections of three corms showing thickness of rotted tissues.

hard, rough, and usually somewhat scaly after the corms are dry. The affected area is sunken, and there is a sharp line of demarcation between diseased and healthy tissues.

While the majority of diseased corms in any given lot usually have symptoms characteristic of only one of the disease forms, it is not uncommon to find corms that have symptoms of two of the disease forms or symptoms intermediate between them, fig. 4. Bald (1953) stated, "In any large collection of gladiolus varieties infected with *Fusarium* diseases it has not been found possible to maintain on a symptomological basis the division between *Fusarium* basal rot and *Fusarium* yellows. On different varieties a gradation was found between the 2 symptom types."

ETIOLOGY

The etiology of the *Fusarium* disease of gladiolus is quite typical for that of plant diseases caused by fungi of the genus *Fusarium*. The occurrence of the disease in more than one form and the great variability commonly found in species of *Fusarium* have contributed to the confusion regarding the cause of this disease.

Previous Accounts

A report by Massey (1922) was the first published account of a gladiolus disease in which a fungus of the genus *Fusarium* was established as the cause. A more extensive description of this disease and its causal agent was later published by

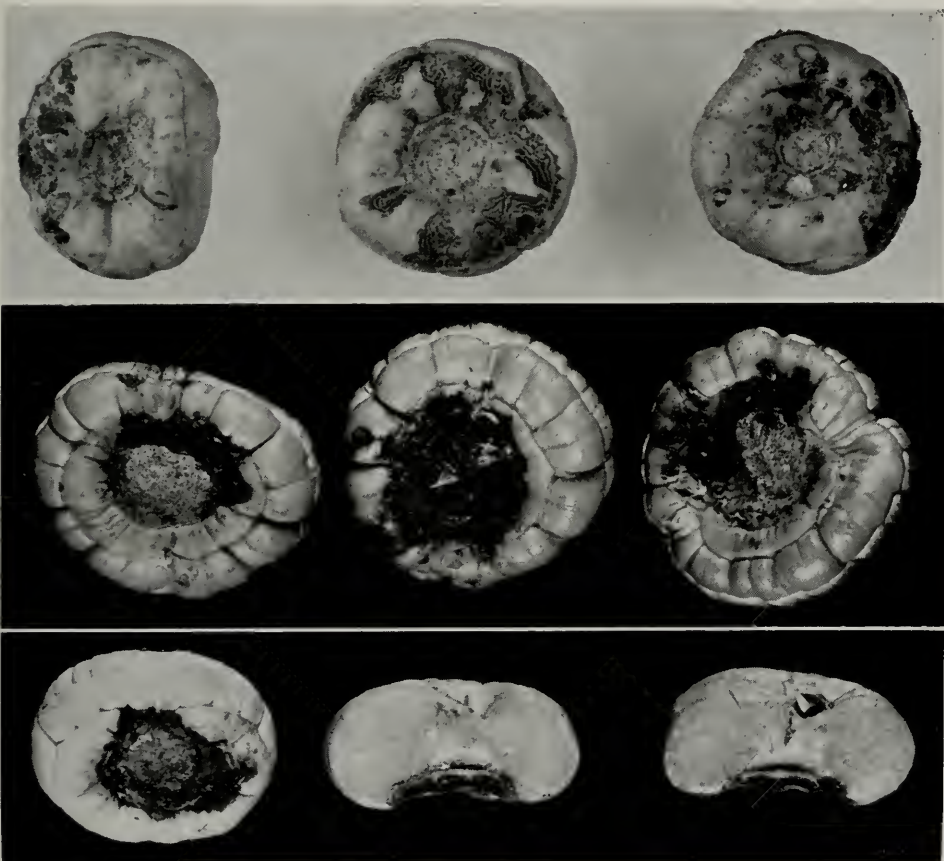


Fig. 3.—Corms of three gladiolus varieties affected with the basal dry rot form of the *Fusarium* disease: top row, variety Gold Eagle; middle row, Lake Placid; bottom row, Spotlight. The two sectioned Spotlight corms show the extreme thinness of the rotted tissue.

the same worker (1926). Massey classified the organism as *Fusarium oxysporum* Schlecht. emend. Wr. var. *gladioli* n. var.

McCulloch (1944) considered the *Fusarium* she found associated with the vascular form of the disease to be sufficiently distinct from *Fusarium oxysporum* var. *gladioli*, described by Massey, to warrant putting it in another species. She classified it as *Fusarium orthoceras* App. et Wr. var. *gladioli*. In comparing the two organisms she stated, "In culture the bulb-rot organism has, in most tests and examinations, shown less abundant aerial growth, less pigment, and wider macrospores than the yellows organism. . . . The most distinctive characteristics of these two *Fusaria* of *gladiolus* are the effects on the host."

Other workers have been inconsistent in

their use of names for the causal agents of the different forms of the disease. McClellan (1945) used the name *Fusarium orthoceras* App. & Wr. var. *gladioli* McCulloch for the vascular *Fusarium* of *gladiolus*. In a later article the same writer (1947) listed *F. oxysporum* f. *gladioli* Sny. & Hans. as the causal agent for yellows and rot. He described the other forms of the disease but did not name the causal agents. McClellan & Stuart (1947) used the name *F. oxysporum* f. *gladioli* (Massey) Sny. & Hans. for the causal agent of *gladiolus* "yellows, or corm rot." McClellan (1948) used both names, *F. oxysporum* var. *gladioli* Massey and *F. orthoceras* var. *gladioli* McCulloch. Nelson (1948) listed *F. orthoceras* Woll. var. *gladioli* McCull. as the cause of yellows,

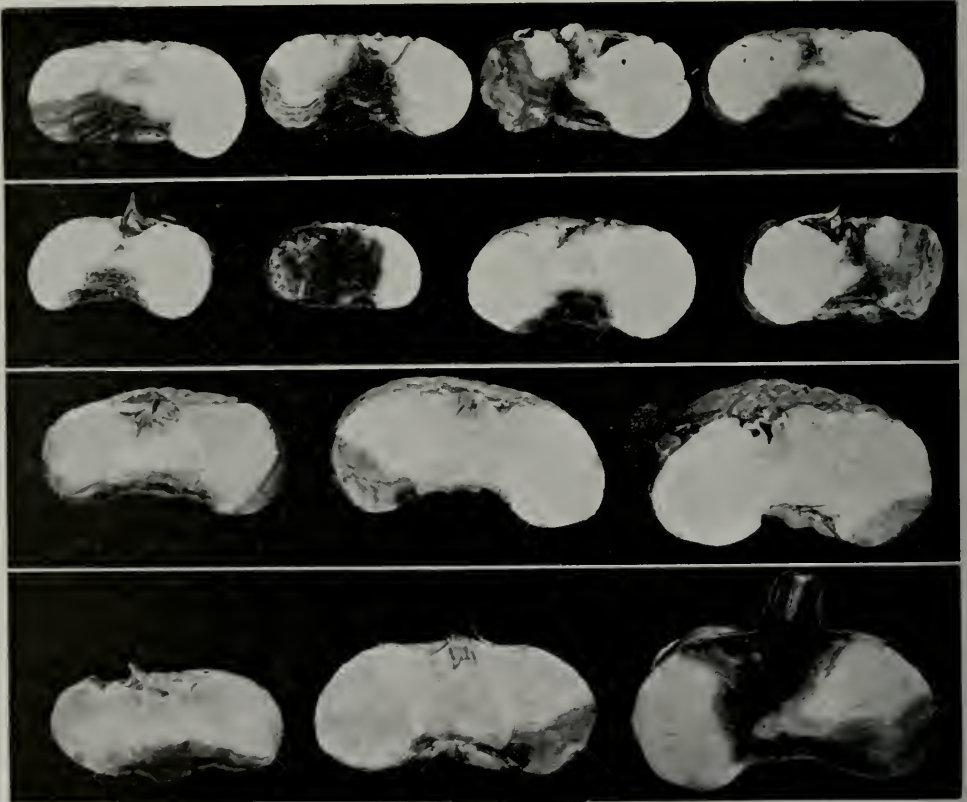


Fig. 4.—Sectioned corms of *gladiolus* variety Dieppe, in upper two rows, and Golden Arrow, in lower two rows, showing symptoms of all three forms of the *Fusarium* disease. The second corm from the left in the top row shows core rot and vascular discoloration. The first and third corms in the second row and the fourth corm in the top row show symptoms intermediate between basal dry rot and core rot. The first corms in the third and fourth rows have basal dry rot. The other corms show the brown rot form of the *Fusarium* disease.

F. oxysporum Schlecht. var. *gladioli* Massey as the cause of brown rot, and merely *Fusarium* sp. as the cause of basal dry rot. Gould (1949) called *F. oxysporum* f. *gladioli* the cause of Fusarium rot. Miller & Magie (1950) listed *F. oxysporum* f. *gladioli* as the cause of Fusarium storage rot of gladiolus corms. Magie (1950) referred to the causal agent of Fusarium yellows and Fusarium corm rot as *F. oxysporum* f. *gladioli*.

Bald (1953) stated, "Typical single spore cultures from basal rot, yellows, and intermediate infections were submitted to Dr. W. C. Snyder for identification. He placed all the cultures in the species *Fusarium oxysporum* Schl. As these Fusaria were obtained from active lesions, and some similar isolates were shown by inoculation tests to be pathogenic on gladiolus corms, the strains causing basal rot, yellows, and intermediate symptoms on gladioli in southern California have been provisionally grouped under the name *F. oxysporum* f. *gladioli* (Massey) Sny. and Hans."

Difficulties in Classifying Fusaria

A review of the literature indicates that the difficulties encountered by plant pathologists in classifying strains of the gladiolus *Fusarium* have been great and are similar to the experiences of many other workers faced with the task of determining the relationship and specific name to be applied to a pathogenic *Fusarium*.

Some of the confusion is a result of the use of two different systems of nomenclature and taxonomy now available for naming and classifying isolates of *Fusarium*. These are the detailed system of Wollenweber & Reinking (1935) and the simplified system of Snyder & Hansen (1940, 1941, 1945).

In the system of Wollenweber & Reinking (1935), the genus *Fusarium* is divided into 16 named sections in which a total of 65 species, 55 varieties, and 22 forms are differentiated. This system is based largely on recommendations made at a conference held in Madison, Wisconsin, in 1924 (Wollenweber *et al.* 1925). According to these recommendations, species and varieties must be distinguished by

morphological characters only. Each species includes groups of individuals that can be distinguished by morphological characters which must be "of such a nature as to be applicable and usable by mycologists in general and which will be most serviceable for practical purposes." Each variety is distinguished by morphological characters of less importance than those used for specific segregation. Groups of individuals differing from the species and the variety only in certain physiological characters are separated as forms. This system failed to meet with unqualified approval because of difficulties that still were encountered by workers in attempting to classify specific isolates of *Fusarium*.

The simplified system of nomenclature and taxonomy proposed by Snyder & Hansen (1940, 1941, 1945) was based on their extensive investigations of the variability shown in culture by species, varieties, and forms of *Fusarium*. Section *Elegans* was the first to be revised according to their concept of species. Simplification was achieved by emending the description of one species, *Fusarium oxysporum* Schl., to agree with the description of section *Elegans* given by Wollenweber (1913). The 10 species, 18 varieties, and 12 forms comprising section *Elegans* of Wollenweber & Reinking (1935) were placed in one species, *Fusarium oxysporum*, on the sole basis of morphology. Twenty-five parasites of the section were made forms of this common species on the basis of pathogenicity alone. Revision of the other sections followed. As a result of the complete revision of the genus, the 16 sections, 65 species, 55 varieties, and 22 forms of *Fusarium* of Wollenweber & Reinking were reduced to 8 species and 34 forms. No varieties were recognized in the system of Snyder & Hansen.

Massey (1926), McCulloch (1944), and Nelson (1948) used the system of Wollenweber & Reinking to designate species names for the gladiolus *Fusarium*. Gould (1949), Miller & Magie (1950), Magie (1950), and Bald (1953) used the system of Snyder & Hansen. McClellan (1945, 1947, 1948) used both systems. Wollenweber's classification was the only one in existence at the time Massey published the results of his investigations.

Both systems were available to workers after 1940.

PURPOSE OF PRESENT INVESTIGATION

This study was undertaken to rectify the confusion appearing in the literature regarding the relationship between the causal agent or agents of the different forms of the *Fusarium* disease on gladiolus and the symptoms produced. The main object of the investigation was to determine if different strains of *Fusarium* produced the different symptoms and if these

strains could be fitted into well-defined groups on the basis of their pathogenicity and physiological characters.

METHODS

Several hundred isolates of *Fusarium* were cultured by the writer from diseased gladiolus corms in the years 1945 through 1950. Thirty-three of these isolates together with six isolates received from Ray Nelson of Michigan State College and one isolate from Robert O. Magie of the University of Florida Gulf Coast Experiment Station (a total of 40 isolates)

Table 1.—Sources of the 40 isolates of the gladiolus *Fusarium* used in the infection experiments and physiological studies reported in this study.

ISOLATE	DATE OF ISOLATION	DISEASE FORM	GLADIOLUS VARIETY	LOCALITY
45-73.....	Dec. 10, 1945	Vascular	Dr. F. E. Bennett	Wichert, Ill.
45-80.....	Dec. 10, 1945	Vascular	Dr. F. E. Bennett	Wichert, Ill.
46-3.....	Dec. 13, 1946	Vascular	Phyllis McQuiston	Wichert, Ill.
46-5.....	Dec. 14, 1946	Vascular	Phyllis McQuiston	Wichert, Ill.
46-9.....	Dec. 17, 1946	Vascular	Bit o' Heaven	Wichert, Ill.
46-14.....	Nov. 1, 1946	Vascular	Unknown	Cairo, Ill.
47-6.....	Jan. 24, 1947	Vascular	Beacon	Wichert, Ill.
47-10.....	Feb. 5, 1947	Vascular	Dr. F. E. Bennett	Wichert, Ill.
49-4.....	Jan. 14, 1949	Vascular	Margaret Beaton	Wichert, Ill.
49-15.....	Jan. 19, 1949	Vascular	Lantana	Champaign, Ill.
49-23.....	Jan. 21, 1949	Vascular	Myrna	Wichert, Ill.
49-30.....	Jan. 21, 1949	Vascular	Dream of Beauty	Wichert, Ill.
49-31.....	Jan. 21, 1949	Vascular	Mother Kadel	Wichert, Ill.
50-6.....	Feb. 15, 1950	Vascular	Yellow Herald	Wichert, Ill.
50-24*.....	April 2, 1945	Vascular	Unknown	East Lansing, Mich.
50-27*.....	Nov. 4, 1949	Vascular	Corona	Oregon
50-28*.....	April 14, 1950	Vascular	Unknown	California
45-8.....	Feb. 5, 1945	Brown rot	Picardy	Wichert, Ill.
45-74.....	Dec. 14, 1945	Brown rot	Picardy	Wichert, Ill.
45-75.....	Dec. 14, 1945	Brown rot	Picardy	Wichert, Ill.
45-78.....	Dec. 10, 1945	Brown rot	Dr. F. E. Bennett	Wichert, Ill.
46-4.....	Dec. 14, 1945	Brown rot	Phyllis McQuiston	Wichert, Ill.
46-12.....	Dec. 16, 1946	Basal dry rot	Aladdin	Wichert, Ill.
47-1.....	Jan. 23, 1947	Brown rot	Picardy	Wichert, Ill.
47-8.....	Jan. 24, 1947	Basal dry rot	Beacon	Wichert, Ill.
47-12.....	Feb. 5, 1947	Brown rot	Corona	Wichert, Ill.
47-19.....	Feb. 13, 1947	Brown rot	Picardy	Fort Collins, Colo.
47-32.....	Sept. 27, 1947	Brown rot	Unknown	Cocoa, Fla.
49-1.....	Jan. 14, 1949	Brown rot	Margaret Beaton	Wichert, Ill.
49-17.....	Jan. 19, 1949	Brown rot	Abu Hassan	Wichert, Ill.
49-19.....	Jan. 19, 1949	Brown rot	Wings of Song	Wichert, Ill.
50-7.....	Feb. 15, 1950	Brown rot	Ohio Nonpareil	Champaign, Ill.
50-22†.....	1950	Brown rot	Spic and Span	Bradenton, Fla.
50-25*.....	Sept. 3, 1948	Brown rot	Picardy	East Lansing, Mich.
47-2.....	Jan. 23, 1947	Basal dry rot	Picardy	Wichert, Ill.
47-3.....	Jan. 24, 1947	Basal dry rot	Picardy	Wichert, Ill.
49-8.....	Jan. 14, 1949	Brown rot	Valeria	Wichert, Ill.
49-20.....	Jan. 19, 1949	Basal dry rot	Wings of Song	Wichert, Ill.
50-23*.....	Nov. 25, 1936	Basal dry rot	Souvenir	East Lansing, Mich.
50-26*.....	Dec. 23, 1948	Basal dry rot	Souvenir	East Lansing, Mich.

*Isolated by Ray Nelson, Michigan State College, East Lansing.

†Isolated by Robert O. Magie, University of Florida Gulf Coast Experiment Station, Bradenton.

Table 2.—Growth of six isolates of the gladiolus *Fusarium* on Coons's agar at various temperatures. Incubation period 160 hours.

TEMPERATURE, DEGREES C.	MEAN OF DIAMETERS (MILLIMETERS) OF TWO COLONIES					
	Brown Rot Isolates		Vascular Isolates		Basal Dry Rot Isolates	
	45-75	50-22	45-73	50-24	47-3	50-23
1-3.....	0.0	0.0	0.0	0.0	0.0	0.0
5.....	0.0*	0.0	0.0	0.0*	0.0	0.0
20-22.....	52.0	63.0	66.0	50.0	58.0	51.5
24.....	67.0	78.0	83.0	63.0	64.0	63.0
26.....	78.0	85.0	90.0	68.0	73.0	74.0
27-28.....	74.0	87.0	90.0	71.0	75.0	75.0
30-31.....	43.5	49.0	57.0	50.0	49.0	49.0
32.....	39.5	48.0	54.0	47.0	48.0	47.0
36.....	5.5	8.5	7.0	11.5	6.0	6.0
37.....	2.5	3.5	3.0	5.5	4.0	3.0
39.....	0.0	0.0	0.0*	0.0	0.0	0.0*
40.....	0.0	0.0	0.0	0.0	0.0	0.0

*A trace of growth insufficient for measurement appeared in these plates.

were selected for comparison in pathogenicity tests and physiological studies. Seventeen of these isolates were obtained from corms with the vascular, 16 from corms with the brown rot, and 7 from corms with the basal dry rot form of the disease. They will be referred to hereafter as vascular, brown rot, and basal dry rot or basal rot isolates.

The original isolations were made on *Difco* potato dextrose agar. Single-spore isolates were obtained from spore suspensions in sterile water blanks; each suspension was diluted until a desired spore concentration was reached, and then the diluted suspension was poured over the surface of a thin film of 2 per cent water agar in a Petri dish. After a few seconds the excess suspension was poured off and the plate was allowed to stand for 15 to 16 hours. The Petri dish was then placed on the stage of a dissecting microscope, and germinated spores were picked off singly on the tip of a needle and transferred to potato dextrose agar slants. Progenies from these single-spore isolates were used in the infection experiments and physiological studies. The sources of all isolates are listed in table 1.

Comparisons of isolates from the three forms of the disease were made on the following bases: reactions to temperature, reactions to aniline dyes, reactions to copper salts, reactions to mercuric chloride, color reactions on steamed rice, growth

types on Wellman's differential medium, pH changes produced in liquid media, spore measurements, inoculation tests in the laboratory, and inoculation tests in the greenhouse.

PHYSIOLOGICAL STUDIES

Physiological studies of the gladiolus *Fusarium* were made by the writer in an effort to determine if isolates from the three forms of the disease could be distinguished by their physiological characters.

Influence of Temperature on Growth Rates

Massey (1926) reported that the gladiolus *Fusarium* studied by him grew in culture at temperatures ranging from 5 degrees to 35 degrees C.; the optimum was 27.5 degrees. McCulloch (1944) reported that the *Fusarium* she studied grew at temperatures ranging from less than 3 degrees to about 34-36 degrees C.; the optimum temperature range was 23-26 degrees C.

In the present investigation, the influence of temperature on the growth rates of the gladiolus *Fusarium* was studied on two isolates grown from each disease type on Coons's agar in Petri dishes through a series of 12 temperatures. The inoculum for the Petri dishes was prepared in the following manner to obviate erratic be-

havior traceable to size or character of inoculum: coarse white cotton thread was cut in pieces about $1\frac{1}{2}$ inches long and autoclaved in distilled water. These pieces were laid on an agar slant, which was then inoculated from the isolate to be tested. About a week after fungus growth had overrun the cotton threads, the threads were lifted from the culture, scraped clean of adhering agar, and cut with sterile scissors into sections, each about 2 millimeters long. One piece of *Fusarium*-infested thread was placed in the center of each dish. Immediately after inoculation

the dishes were placed in electrically controlled incubators kept at temperatures shown in table 2 and left for 160 hours. At the end of the incubation period the diameters of the colonies were measured, table 2. Two dishes of each isolate were used for each temperature. Relative sizes of the colonies grown at different temperatures are shown in figs. 5, 6, 7, and 8.

None of the isolates grew at the 1-3 degree C. temperature range, table 2, and only brown rot isolate 45-75 and vascular isolate 50-24 showed traces of growth at 5 degrees C. Since the next higher tempera-

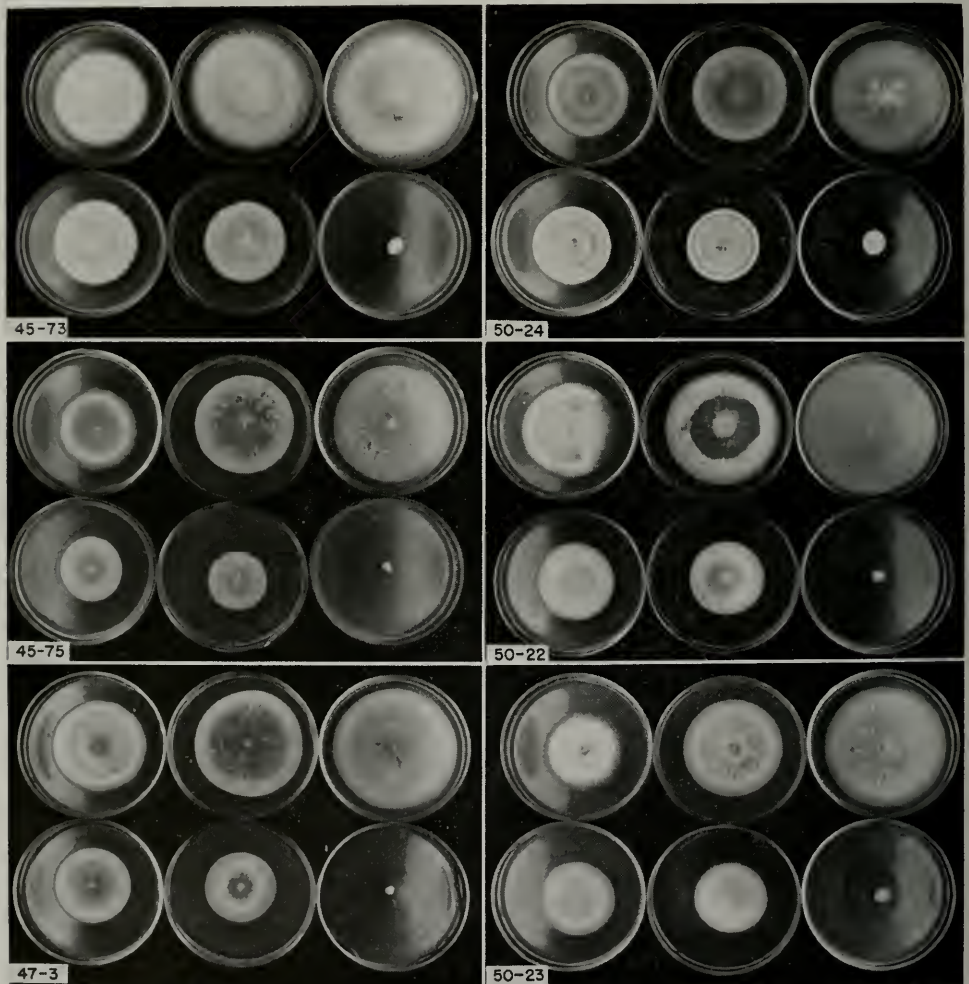


Fig. 5.—Six isolates of *Fusarium* grown on Coons's agar for 160 hours at the following temperatures: 20-22, 24, 27-28 degrees C. (top row in each set of six dishes, left to right); 30, 32, and 36 degrees C. (bottom row in each set, left to right). Isolates 45-73 and 50-24 are vascular isolates, 45-75 and 50-22 are brown rot isolates, 47-3 and 50-23 are basal dry rot isolates.

ture range used was 20–22 degrees C., the minimum temperature for growth of these isolates was not determined. The range of temperatures for optimum growth of all six isolates was 26–28 degrees C. All isolates grew at 37 degrees C., but only vas-

cular isolate 45-73 and basal dry rot isolate 50-23 showed traces of growth at 39 degrees C. None of the isolates grew at 40 degrees C. After the test period, all Petri dishes that had been incubated at 40 degrees were kept at room temperature for

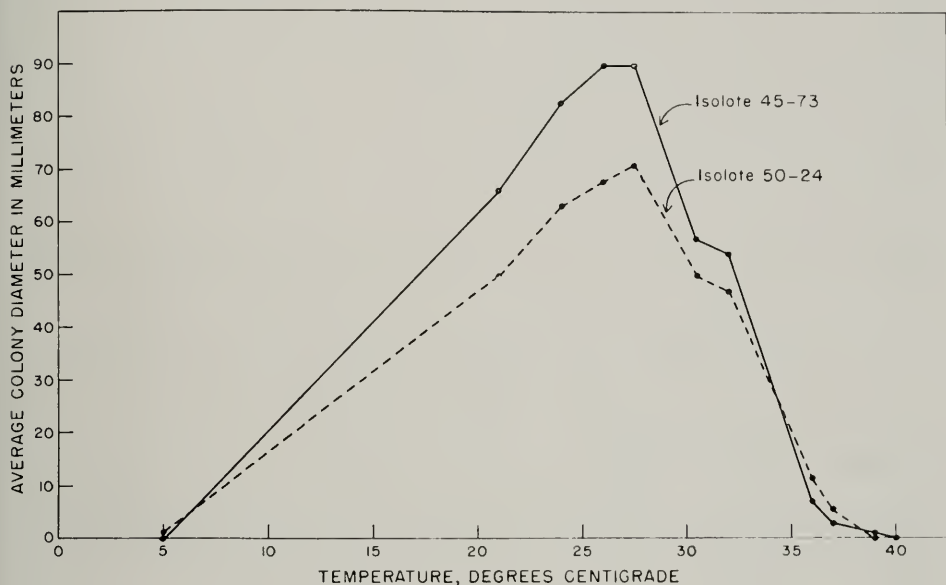


Fig. 6.—Effect of temperature on the colony size of two vascular isolates of *Fusarium* grown 160 hours on Coons's agar.

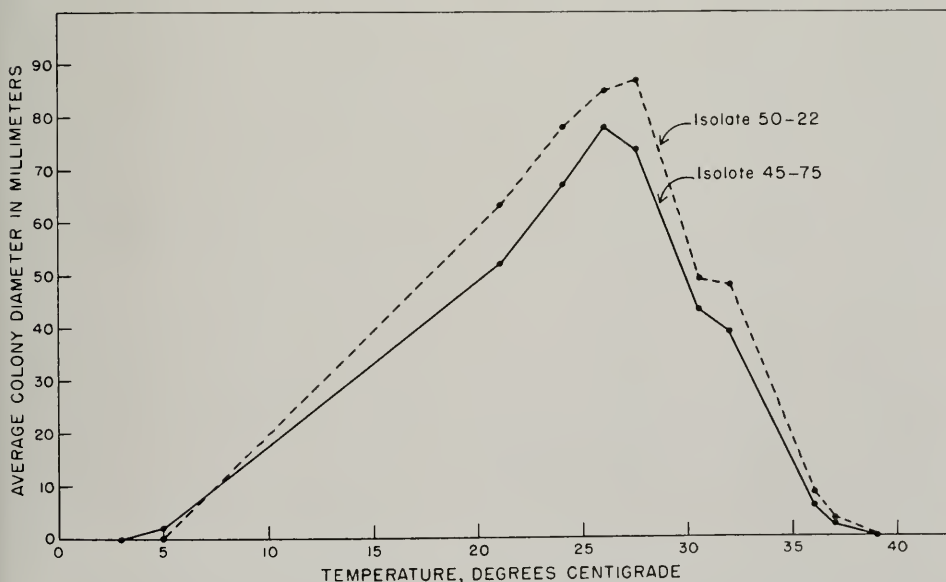


Fig. 7.—Effect of temperature on the colony size of two brown rot isolates of *Fusarium* grown 160 hours on Coons's agar.

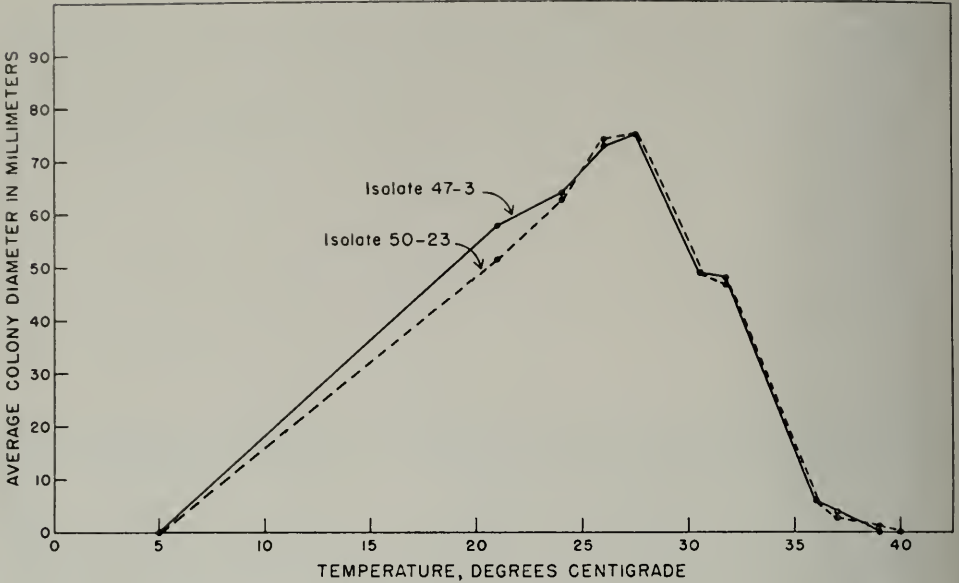


Fig. 8.—Effect of temperature on the colony size of two basal dry rot isolates of *Fusarium* grown 160 hours on Coons's agar.

several days. Five of the six isolates grew when moved to the lower temperature. Basal dry rot isolate 47-3 apparently had been killed by the 160-hour exposure to 40 degrees C.

In these studies the general shape of the curves of growth responses of all isolates to various temperatures was of the same general pattern, figs. 6, 7, and 8. The basal dry rot isolates agreed much more closely in their responses to temperatures than did the isolates from the other disease forms. The vascular isolates showed the greatest divergence in responses to temperatures. Isolates from the three disease forms could not be distinguished by their growth responses to various temperatures.

Reactions to Aniline Dyes

The 40 isolates of the gladiolus *Fusarium* listed in table 1 were grown on Coons's agar containing various concentrations of the aniline dyes malachite green, brilliant green, and crystal violet, according to the method developed by Coons & Strong (1931).

The malachite green and brilliant green media were made from preparations of stock agar containing 10 milliliters of 0.5

per cent dye solution per liter of Coons's agar. For each medium, four concentrations, tables 3 and 5, were prepared from the stock agar diluted with appropriate amounts of sterile, melted Coons's agar.

The crystal violet stock agar was prepared from 100 milliliters of 0.5 per cent dye solution added to a liter of Coons's agar, and the four concentrations, table 4, were made from this stock.

Inoculum was prepared in the manner described for the temperature studies. Four isolates were grown in each dish. The center of each quadrant of the dish was seeded with a 2-millimeter piece of *Fusarium*-infested thread. The cultures were run in duplicate and were incubated for 10 days at 25 degrees C.

Readings of the Petri dish cultures were made in accordance with the decimal numbering scheme used by Coons & Strong (1931). This scheme is as follows:

Color of mycelium	
White	10.
Red	20.
Breadth of mycelial mat	
No growth	0
Very slight growth	tr.
.5 cm. to 1 cm.	1.
1 cm. to 2 cm.	2.

2 cm. to 3 cm.	3.
3 cm. to 4 cm.	4.
4 cm. to 5 cm.	5.
Changes in medium color	
No change0
Halo1
Strong decolorization2
Color intensified in mycelium3
Edge of colony	
Even00
Ramose01
Frondose02
Growth form of mycelium	
Submerged000

Cottony001
Villous002
Sericeous003
Tufted004
Submerged, cottony center005
Submerged, sericeous center006
Submerged, with cottony fringe at edge, atoll....	.007
Woolly008

In the key to the species of *Fusarium* as arranged by Coons & Strong (1931), *F. orthoceras* and *F. oxysporum* are recorded as being moderately sensitive to malachite green, i.e., growth usually 1-2 centime-

Table 3.—Reactions* of isolates of the gladiolus *Fusarium* to four concentrations of malachite green in Coons's agar.

ISOLATE	CONCENTRATION OF DYE				
	1:66,666	1:100,000	1:200,000	1:400,000	Control
45-73.....	0	tr	11.001	12.001	15.001
45-80.....	tr	tr	11.001	12.001	15.007
46-3.....	0	tr	11.001	13.001	15.000
46-5.....	0	tr	11.001	12.001	15.000
46-9.....	0	tr	11.001	12.001	15.007
46-14.....	0	tr	11.001	12.001	15.000
47-6.....	0	tr	12.001	12.001	15.001
47-10.....	0	0	tr	12.001	15.000
49-4.....	tr	11.001	12.001	14.003	15.007
49-15.....	0	0	tr	12.001	15.007
49-23.....	tr	11.001	12.001	15.001	15.004
49-30.....	tr	0	tr	12.014	15.007
49-31.....	0	tr	11.001	12.004	15.000
50-6.....	0	tr	11.001	13.001	15.001
50-24.....	tr	11.001	12.001	13.001	15.000
50-27.....	tr	tr	11.001	12.001	15.007
50-28.....	0	tr	11.001	12.001	15.007
45-8.....	0	0	11.001	13.001	15.001
45-74.....	0	tr	11.014	13.004	15.005
45-75.....	0	tr	11.001	13.001	15.001
45-78.....	0	0	0	12.001	15.001
46-4.....	0	tr	11.014	12.004	15.004
46-12.....	0	tr	tr	11.001	15.000
47-1.....	0	tr	12.004	14.003	15.004
47-8.....	0	tr	12.001	14.003	15.001
47-12.....	0	tr	11.001	12.001	15.000
47-19.....	0	tr	11.001	13.004	15.000
47-32.....	0	tr	11.001	12.001	15.001
49-1.....	0	tr	12.001	14.005	15.000
49-17.....	0	tr	11.004	12.012	15.001
49-19.....	0	tr	11.001	12.001	15.001
50-7.....	0	tr	12.001	14.001	15.001
50-22.....	tr	tr	12.004	14.005	15.001
50-25.....	0	tr	11.005	12.001	15.000
47-2.....	tr	tr	12.004	14.002	15.000
47-3.....	0	tr	11.005	12.001	15.001
49-8.....	0	0	11.001	12.001	15.007
49-20.....	0	tr	11.001	0	15.001
50-23.....	0	tr	11.001	12.001	15.007
50-26.....	0	tr	tr	12.001	15.001

*Reactions are expressed in the numbering scheme of Coons & Strong (1931), as explained in the text.

ters broad in the 1:400,000 and 1:200,000 concentrations. Growth on crystal violet extended to about 1:5,000 or 1:4,000. At this point in the key the two species are separated by their reactions to brilliant green, brilliant green not being decolorized by *F. orthoceras* but being decolorized by *F. oxysporum*. Also, a difference in growth form of the two species on crystal violet is noted, growth being submerged with sericeous center for *F. orthoceras* and cottony for *F. oxysporum*.

Diameters of the mycelial mats produced in different concentrations of dye

were used by the writer to compare abilities of isolates to tolerate the dye in the culture medium. Considerable variation in diameters was shown by the 40 isolates of gladiolus *Fusarium*, tables 3, 4, and 5. Although of little diagnostic value, growth forms of the various isolates were probably the most striking characters, figs. 9 and 10.

When these isolates were taken through Coons & Strong's key, 26 keyed out to *Fusarium orthoceras* var. *triseptatum*. Since no isolates decolorized brilliant green, they did not fall into *F. oxysporum*. Vascular isolates 49-4 and 49-23, brown

Table 4.—Reactions* of isolates of the gladiolus *Fusarium* to four concentrations of crystal violet in Coons's agar.†

ISOLATE	CONCENTRATION OF DYE			
	1:2,000	1:5,000	1:10,000	1:20,000
45-73.....	tr	11.011	11.011	11.001
45-80.....	0	11.001	12.001	11.000
46-3.....	0	11.001	12.001	12.001
46-5.....	0	11.004	12.000	12.000
46-9.....	0	11.001	12.001	12.001
46-14.....	0	11.001	12.001	12.001
47-6.....	0	11.001	12.001	12.001
47-10.....	0	11.001	12.001	12.001
49-4.....	tr	11.001	12.001	12.011
49-15.....	0	11.001	11.001	11.001
49-23.....	0	12.001	12.001	12.001
49-30.....	tr	11.014	12.001	12.014
49-31.....	tr	11.001	12.001	12.001
50-6.....	0	11.001	11.011	11.001
50-24.....	tr	11.001	11.001	11.001
50-27.....	tr	11.001	12.001	12.001
50-28.....	0	11.001	12.001	12.001
45-8.....	0	12.001	12.001	12.001
45-74.....	11.001	12.014	12.014	12.014
45-75.....	11.001	11.001	12.001	12.001
45-78.....	0	12.001	12.001	12.001
46-4.....	11.001	12.014	12.014	12.014
46-12.....	tr	11.001	12.001	12.001
47-1.....	tr	11.014	12.014	12.014
47-8.....	tr	11.001	12.001	12.001
47-12.....	tr	11.007	11.007	12.007
47-19.....	0	11.014	12.014	12.014
47-32.....	0	11.001	11.001	11.001
49-1.....	11.007	11.007	12.001	12.001
49-17.....	0	12.001	12.000	12.001
49-19.....	11.004	12.004	12.014	12.004
50-7.....	tr	12.014	12.014	12.014
50-22.....	tr	12.004	12.004	12.004
50-25.....	tr	11.000	11.000	11.000
47-2.....	11.001	12.004	12.004	12.004
47-3.....	tr	12.001	12.001	12.001
49-8.....	tr	11.001	12.001	12.001
49-20.....	tr	11.001	12.001	12.001
50-23.....	tr	11.001	11.000	11.000
50-26.....	tr	11.001	11.001	11.001

*Reactions are expressed in the numbering scheme of Coons & Strong (1931), as explained in the text.

†See table 3 for control readings.

rot isolates 47-1, 50-7, and 50-22, and basal dry rot isolate 47-8 keyed out to *F. euoxysporum*. Brown rot isolates 45-74, 45-75, 46-4, and 49-19 fell into *F. orthoceras* var. *longius*, and brown rot isolate 49-1 and basal dry rot isolate 47-2 keyed out to *F. moniliforme*. Basal dry rot isolates 46-12 and 49-20 keyed out to *F. filiferum*, but according to Coons & Strong (1931) this species decolorizes crystal violet. None of the isolates of the gladiolus *Fusarium* decolorized crystal violet.

Although, on the basis of their reactions to the three aniline dyes, the majority of

the isolates of gladiolus *Fusarium* fell into the *F. oxysporum*-*F. orthoceras* group, reactions of so many of the isolates varied from typical reactions of this group that it would be impossible to use the aniline dye method of classifying *Fusaria* from gladiolus with any degree of certainty. Neither would it be possible to separate isolates of the three disease forms, vascular, brown rot, and basal dry rot, on the basis of their reactions to aniline dyes.

The results of this phase of the investigation agree with the results of Moore & Chupp (1952), who found that certain

Table 5.—Reactions* of isolates of the gladiolus *Fusarium* to four concentrations of brilliant green in Coons's agar.†

ISOLATE	CONCENTRATION OF DYE			
	1:25,000	1:50,000	1:100,000	1:200,000
45-73.....	0	tr	12.001	12.301
45-80.....	0	0	12.024	13.004
46-3.....	0	tr	12.001	13.001
46-5.....	0	0	11.001	12.001
46-9.....	0	tr	12.001	13.001
46-14.....	0	0	12.001	13.004
47-6.....	0	0	12.001	13.001
47-10.....	0	0	12.001	12.001
49-4.....	0	12.001	14.001	15.001
49-15.....	0	0	12.001	13.001
49-23.....	0	12.001	14.001	14.001
49-30.....	0	0	12.014	13.001
49-31.....	0	0	11.001	13.001
50-6.....	0	0	12.001	13.001
50-24.....	11.001	12.001	13.001	14.001
50-27.....	0	11.001	12.001	13.001
50-28.....	0	tr	12.001	13.001
45-8.....	0	11.001	12.001	13.001
45-74.....	0	0	12.004	13.004
45-75.....	0	tr	12.001	13.001
45-78.....	0	0	tr	11.011
46-4.....	0	11.014	12.014	13.001
46-12.....	0	tr	11.001	12.001
47-1.....	0	11.014	13.014	14.014
47-8.....	0	11.014	13.001	14.001
47-12.....	0	tr	12.001	13.301
47-19.....	0	11.004	13.024	14.024
47-32.....	0	tr	11.001	12.001
49-1.....	0	11.001	12.005	13.005
49-17.....	0	12.014	13.014	14.004
49-19.....	0	12.004	12.014	13.004
50-7.....	0	12.011	13.011	14.024
50-22.....	0	11.011	12.005	14.005
50-25.....	0	tr	11.001	13.001
47-2.....	0	11.004	13.004	14.014
47-3.....	0	11.001	12.301	13.301
47-8.....	0	11.014	12.001	13.004
49-8.....	0	11.014	12.001	13.001
49-20.....	0	11.001	12.301	13.001
50-23.....	0	tr	11.001	12.001
50-26.....	0	tr	11.001	12.001

*Reactions are expressed in the numbering scheme of Coons & Strong (1931), as explained in the text.

†See table 3 for control readings.

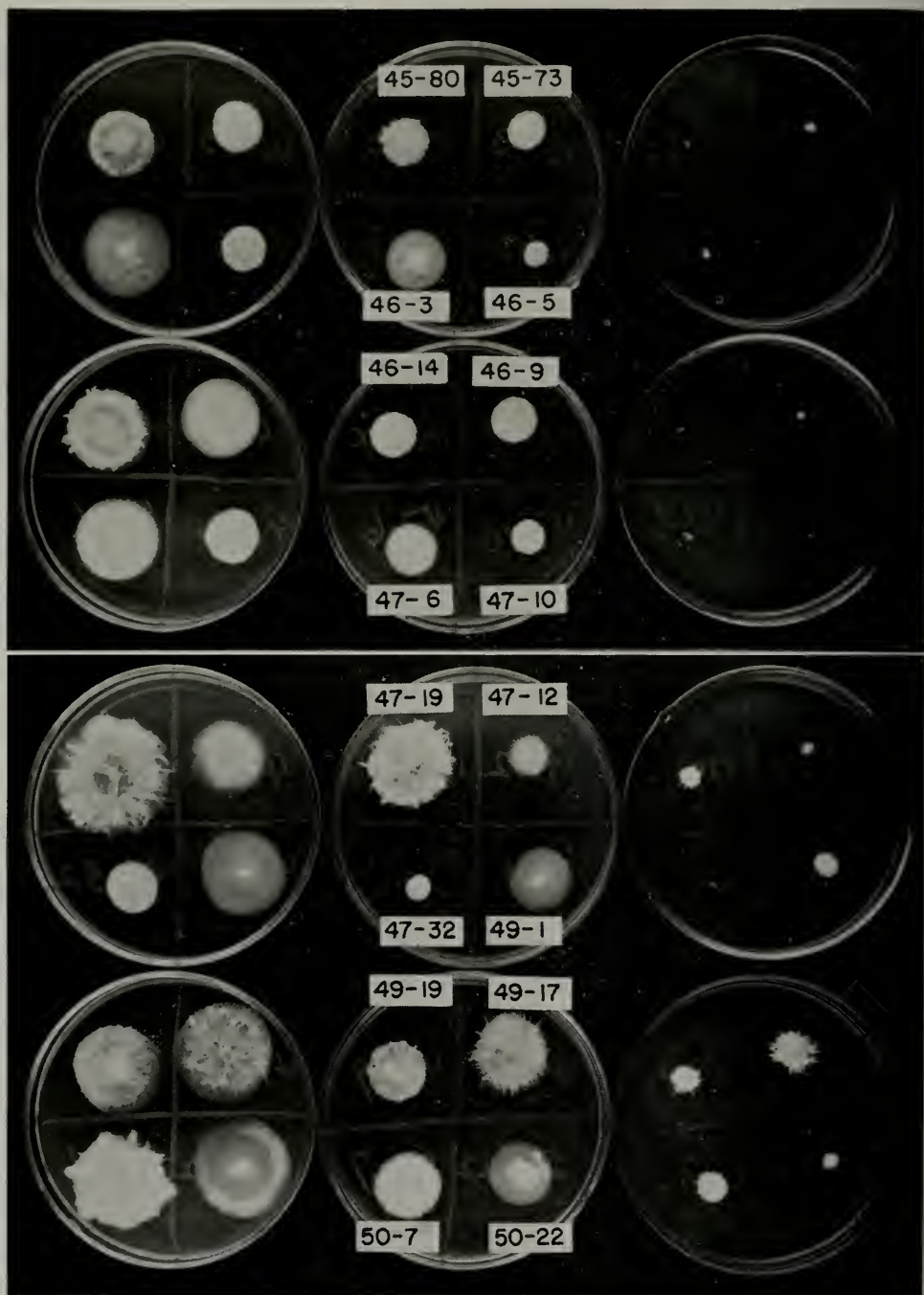


Fig. 9.—Isolates of *Fusarium* grown 10 days on Coons's agar containing three concentrations of brilliant green: left to right 1:200,000, 1:100,000, 1:50,000. Vascular isolates 45-73, 45-80, 46-3, 46-5, 46-9, 46-14, 47-6, and 47-10 are in the two top rows of dishes; brown rot isolates 47-12, 47-19, 47-32, 49-1, 49-17, 49-19, 50-7, and 50-22 are in the two lower rows. Isolates in the dishes at right and left are in the same relative positions as those in the center dishes. Other isolates in this series are shown in fig. 10.

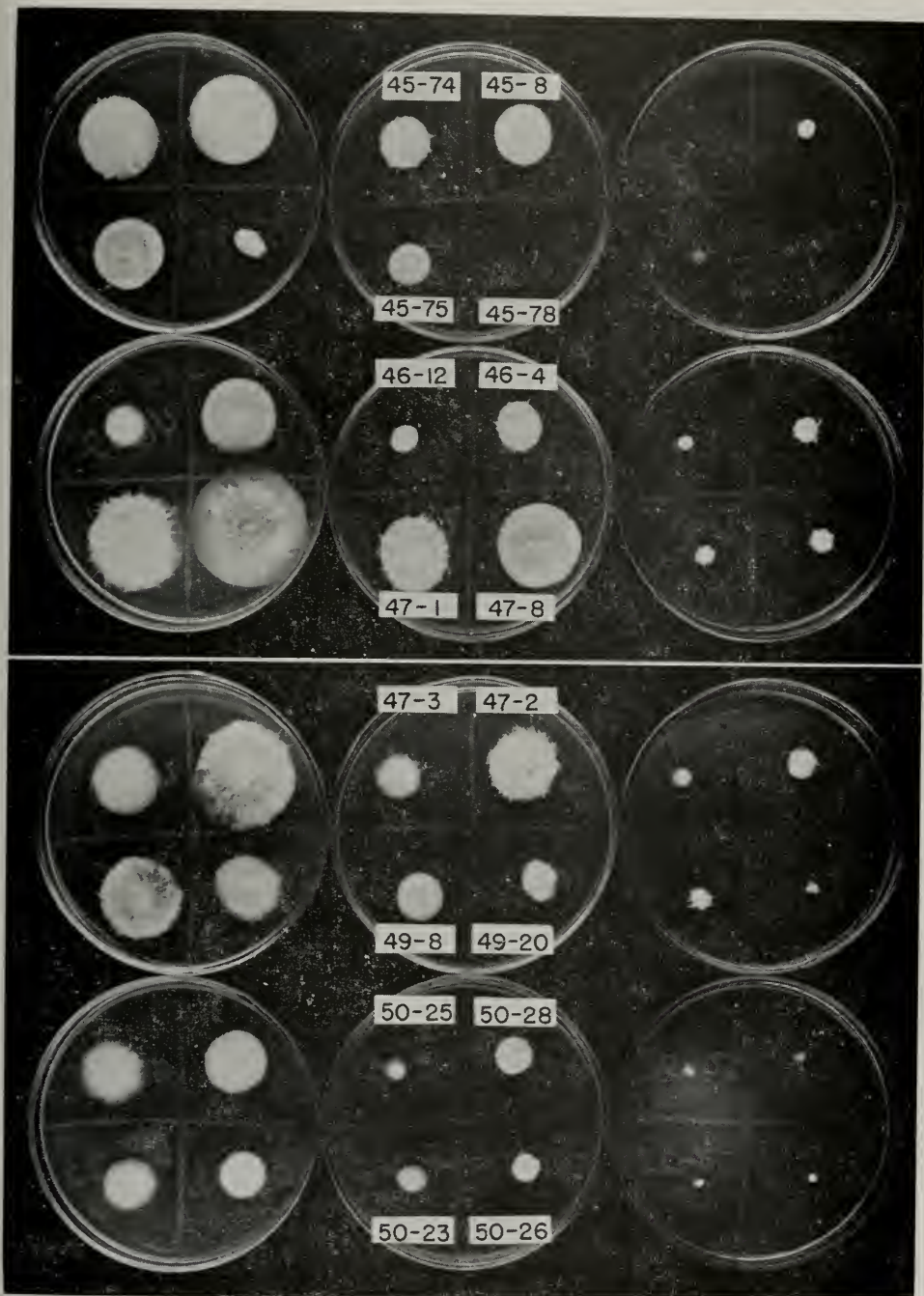


Fig. 10.—Isolates of *Fusarium* grown 10 days on Coon's agar containing the same concentrations of brilliant green as those shown in fig. 9: top row, four brown rot isolates; second row, brown rot isolates 46-4 and 47-1 and basal dry rot isolates 46-12 and 47-8; third row, basal dry rot isolates 47-2, 47-3, and 49-20 and brown rot isolate 49-8; bottom row, basal dry rot isolates 50-23 and 50-26, brown rot isolate 50-25, and vascular isolate 50-28. Isolates in the dishes at right and left are in the same relative positions as those in the center dishes.

isolates of *Fusarium* causing wilts of tomato, cabbage, and muskmelon did not react toward malachite green and crystal violet in the way described by Coons & Strong (1931) for those species.

Reactions to Copper Salts

Coons & Strong (1931) used copper sulfate in the culture medium in some preliminary tests and expressed the opinion that this salt might be useful in identifying some species of *Fusarium*. Nelson, Coons, & Cochran (1937) reported that two

forms of *Fusarium* which cause two forms of the yellows disease of celery could be differentiated if isolates of them were grown on synthetic agar containing either copper sulfate or copper chloride.

The 40 isolates of gladiolus *Fusarium* used in the investigation reported here were grown on Coons's agar to which amounts of copper sulfate were added to make a series of concentrations that ranged from 1/100 molar (*M*/100) to 1/7000 molar, table 6. The strongest concentration, *M*/100, was prepared from 2.5 grams of copper sulfate (1 mole = 249.6

Table 6.—Reactions* of isolates of the gladiolus *Fusarium* to seven different molar (*M*) concentrations of copper sulfate in Coons's agar.†

ISOLATE	CONCENTRATION OF COPPER SULFATE						
	<i>M</i> /100	<i>M</i> /500	<i>M</i> /1000	<i>M</i> /3000	<i>M</i> /5000	<i>M</i> /6000	<i>M</i> /7000
45-73.....	0	0	tr	12.301	—	—	—
45-80.....	0	0	tr	tr	13.304	14.304	15.004
46-3.....	tr	11.329	12.301	13.001	—	—	—
46-5.....	0	0	tr	12.304	14.304	14.304	15.000
46-9.....	0	0	0	tr	11.301	13.301	15.001
46-14.....	0	0	0	tr	11.305	12.300	15.005
47-6.....	0	0	0	tr	12.305	13.305	14.001
47-10.....	0	0	0	12.301	14.305	15.005	15.001
49-4.....	tr	12.301	15.001	15.001	—	—	—
49-15.....	0	0	0	0	tr	11.000	14.000
49-23.....	0	tr	13.301	15.001	—	—	—
49-30.....	0	0	0	0	tr	12.000	15.001
49-31.....	tr	12.304	14.001	15.001	—	—	—
50-6.....	0	0	0	tr	13.001	13.301	14.301
50-24.....	0	0	0	14.001	—	—	—
50-27.....	tr	12.301	14.001	15.001	—	—	—
50-28.....	0	0	0	tr	11.300	12.305	14.305
45-8.....	0	0	0	0	tr	12.301	15.001
45-74.....	11.309	13.301	15.001	15.001	—	—	—
45-75.....	tr	12.301	14.301	14.000	—	—	—
45-78.....	tr	12.301	13.301	13.301	—	—	—
46-4.....	tr	12.301	14.001	15.001	—	—	—
46-12.....	0	0	0	tr	tr	12.000	13.000
47-1.....	tr	12.301	13.001	15.001	—	—	—
47-8.....	0	tr	12.001	14.001	—	—	—
47-12.....	tr	13.301	13.301	15.001	—	—	—
47-19.....	tr	12.301	13.301	15.001	—	—	—
47-32.....	0	0	0	tr	tr	12.000	13.000
49-1.....	tr	13.301	13.301	15.001	—	—	—
49-17.....	tr	12.301	14.001	15.001	—	—	—
49-19.....	0	0	0	tr	15.001	15.001	15.001
50-7.....	0	0	0	tr	15.001	15.001	15.001
50-22.....	tr	13.004	15.004	15.004	—	—	—
50-25.....	0	0	0	13.311	15.001	15.001	15.001
47-2.....	0	12.301	14.001	15.001	—	—	—
47-3.....	tr	12.301	14.001	15.001	—	—	—
49-8.....	0	0	0	tr	15.005	15.005	15.005
49-20.....	0	0	0	11.001	15.001	15.005	15.005
50-23.....	0	0	0	tr	tr	tr	15.001
50-26.....	0	0	0	tr	tr	tr	15.001

* Except for .009 (clumpy) of author, reactions are expressed in the numbering scheme of Coons & Strong (1931).
 † See table 3 for control readings.

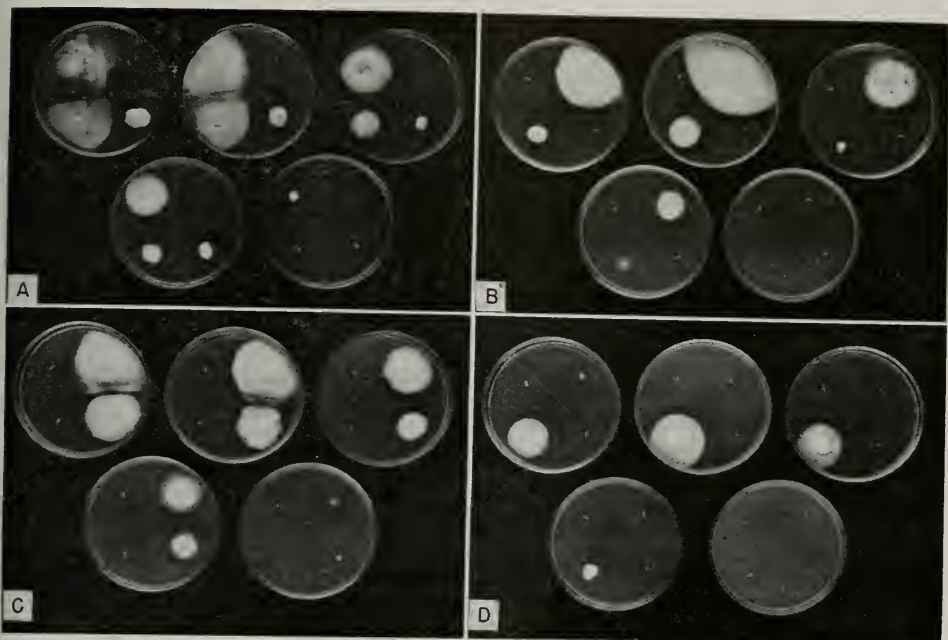


Fig. 11.—Isolates of *Fusarium* grown 10 days in plates of Coons's agar containing (upper rows, left to right: *M*/900, *M*/700, *M*/500, (lower rows, left to right) *M*/300, and *M*/100, copper sulfate. In each plate, clockwise from upper left, isolates are as follows: *A*, brown rot 45-74, 45-8, 45-78, 45-75; *B*, vascular 49-15, 49-4, 49-30, 49-23; *C*, vascular 50-6, 49-31, 50-27, 50-24; *D*, vascular 45-80, 45-73, 46-5, 46-3.

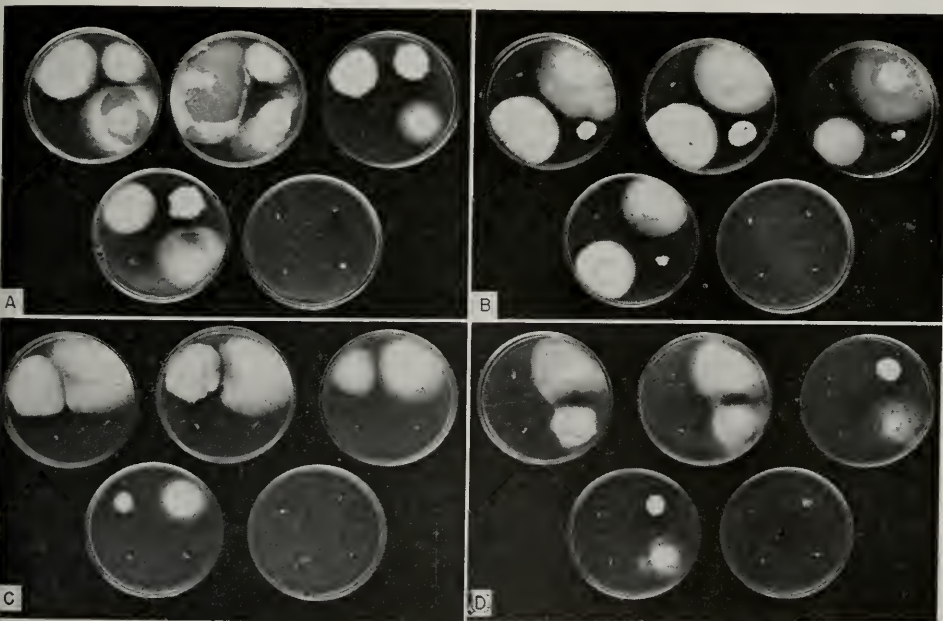


Fig. 12.—As for fig. 11, except *A*, brown rot 47-19, 47-12, 49-1, 47-32; *B*, basal dry rot 46-12, brown rot 46-4, basal dry rot 47-8, brown rot 47-1; *C*, basal dry rot 47-3, 47-2, 49-20, brown rot 49-8; *D*, brown rot 49-19, 49-17, 50-22, 50-7.

grams) dissolved in a liter of Coons's agar. The other concentrations were prepared from this stock diluted with Coons's agar. All dilutions were made immediately after the agar had been autoclaved, and the Petri dishes were poured immediately after the dilutions had been made. This procedure was necessary because it was found that Coons's agar containing copper salts, if remelted after being allowed to solidify, would not resolidify. Inoculum was prepared and the Petri dishes were inoculated in the same manner as that described for the aniline dye tests.

The reactions of the 40 isolates of *gladiolus Fusarium* to copper sulfate are very interesting because they show extreme variability in tolerance to the salt, figs. 11 and 12. One isolate, 45-74, produced measurable growth in the *M*/100 concentration. Seven isolates produced only traces of growth in the *M*/5000 concentration and two isolates produced measurable growth in only the *M*/7000 concentration, table 6. Although the reactions of the various isolates to copper sulfate failed to distinguish between isolates of *Fusarium* from the three disease forms,

Table 7.—Reactions* of isolates of the *gladiolus Fusarium* to four different molar (*M*) concentrations of copper chloride in Coons's agar.†

ISOLATE	CONCENTRATION OF COPPER CHLORIDE			
	<i>M</i> /100	<i>M</i> /300	<i>M</i> /500	<i>M</i> /1000
45-73.....	0	0	0	0
45-80.....	0	0	0	0
46-3.....	11.301	12.101	12.301	12.301
46-5.....	0	0	0	tr
46-9.....	0	0	0	0
46-14.....	0	0	0	0
47-6.....	0	0	0	0
47-10.....	0	0	0	tr
49-4.....	tr	14.301	15.001	15.001
49-15.....	0	0	0	0
49-23.....	0	tr	11.301	15.001
49-30.....	0	0	0	0
49-31.....	13.301	14.301	15.001	15.001
50-6.....	0	0	0	tr
50-24.....	0	0	0	11.000
50-27.....	tr	14.301	15.001	15.001
50-28.....	0	0	0	tr
45-8.....	0	0	0	0
45-74.....	12.301	15.301	15.301	15.001
45-75.....	tr	13.301	14.301	15.001
45-78.....	0	11.301	12.301	13.301
46-4.....	0	14.301	15.001	15.001
46-12.....	0	0	0	0
47-1.....	0	13.301	13.301	15.001
47-8.....	0	tr	13.301	15.001
47-12.....	0	13.311	14.001	15.001
47-19.....	0	15.001	15.001	15.001
47-32.....	0	0	0	0
49-1.....	tr	15.001	15.001	15.001
49-17.....	tr	13.301	15.301	15.001
49-19.....	0	0	0	0
50-7.....	0	0	0	0
50-22.....	11.301	15.301	15.301	15.001
50-25.....	0	0	0	tr
47-2.....	tr	15.301	15.001	15.001
47-3.....	tr	13.301	14.001	15.001
49-8.....	0	0	0	0
49-20.....	0	0	0	0
50-23.....	0	0	0	0
50-26.....	0	0	0	0

*Reactions are expressed in the numbering scheme of Coons & Strong (1931), as explained in the text.

†See table 3 for control readings.

the brown rot isolates were, in general, less sensitive to copper sulfate than were the vascular and basal dry rot isolates.

Copper chloride was used in a way similar to that in which copper sulfate was used. The *M*/100 concentration was prepared from 1.7 grams of copper chloride dissolved in a liter of Coons's agar and the other concentrations were prepared from this stock diluted with Coons's agar. Only four concentrations, *M*/100, *M*/300, *M*/500, and *M*/1000, were used, table 7. Further dilutions of copper chloride were not used because the reactions of the iso-

lates of *Fusarium* appeared to be following the same pattern with copper chloride as they did with copper sulfate.

Reactions to Mercuric Chloride

The preplanting treatment of gladiolus corms in a solution of mercuric chloride has been one of the methods used by growers to control the *Fusarium* disease. Because seemingly erratic results have sometimes been obtained from chemical treatment of corms, it was decided to test the 40 isolates of gladiolus *Fusarium* for sen-

Table 8.—Reactions* of isolates of the gladiolus *Fusarium* to six concentrations of mercuric chloride in Coons's agar.†

ISOLATE	CONCENTRATION OF MERCURIC CHLORIDE					
	1:10,000	1:12,500	1:14,285	1:16,666	1:20,000	1:25,000
45-73.....	0	0	0	0	13.001	14.001
45-80.....	0	0	0	0	15.001	15.000
46-3.....	0	0	15.001	15.001	15.001	15.001
46-5.....	0	0	0	0	15.001	15.001
46-9.....	0	0	0	0	14.001	15.001
46-14.....	0	0	0	0	15.001	15.001
47-6.....	0	0	0	0	15.001	15.001
47-10.....	0	0	0	0	15.001	15.001
49-4.....	0	0	13.001	15.001	15.001	15.001
49-15.....	0	0	0	0	14.001	15.001
49-23.....	0	0	0	0	15.001	15.001
49-30.....	0	0	0	0	14.001	15.001
49-31.....	0	0	0	15.000	15.000	15.000
50-6.....	0	0	0	0	0	14.001
50-24.....	0	12.005	15.005	15.000	15.000	15.000
50-27.....	0	13.001	15.001	15.001	15.001	15.001
50-28.....	0	0	0	0	14.001	15.001
45-8.....	0	0	0	0	0	15.001
45-74.....	0	0	15.001	15.001	15.000	15.001
45-75.....	0	0	15.001	15.001	15.000	15.001
45-78.....	0	0	0	15.001	15.000	15.000
46-4.....	0	13.001	15.001	15.001	15.001	15.001
46-12.....	0	0	0	0	13.001	14.001
47-1.....	0	0	15.001	15.001	15.001	15.001
47-8.....	0	0	0	15.001	15.001	15.000
47-12.....	0	13.001	14.001	14.001	15.001	15.001
47-19.....	0	15.001	15.001	15.001	15.001	15.001
47-32.....	0	0	0	0	13.001	15.001
49-1.....	0	0	0	15.000	15.000	15.000
49-17.....	0	0	15.001	15.001	15.000	15.000
49-19.....	0	0	tr	tr	15.001	15.001
50-7.....	0	0	0	0	13.001	15.001
50-22.....	0	0	15.001	15.001	15.001	15.001
50-25.....	0	0	0	12.001	14.001	15.001
47-2.....	0	0	15.001	15.001	15.001	15.000
47-3.....	0	13.001	14.001	14.001	15.001	15.000
49-8.....	0	0	0	15.001	15.000	15.000
49-20.....	0	0	0	0	13.001	15.000
50-23.....	0	0	0	0	14.001	15.001
50-26.....	0	0	0	12.001	14.001	15.001

*Reactions are expressed in the numbering scheme of Coons & Strong (1931), as explained in the text.

†See table 3 for control readings.

sitivity to various concentrations of mercuric chloride in the culture medium.

A graded series of concentrations of mercuric chloride of 1:1,000 to 1:25,000 was prepared in a manner similar to that used in the tests with copper salts. Results of the test are shown in table 8. Because none of the isolates grew in concentrations of 1:1,000 to 1:10,000, only the dilutions of 1:10,000 and above are shown in table 8.

As in the tests with aniline dyes and copper salts, the isolates varied considerably in their sensitivity to mercuric chloride. One noticeable difference between

the reactions to mercuric chloride and the reactions to the other growth-inhibiting substances was observed. In the various concentrations of aniline dyes and copper salts, size of the colonies of *Fusarium* increased as the dilution became greater. Although this reaction occurred to some extent with mercuric chloride, it was not so pronounced. In most cases, if growth occurred at all it was very vigorous and covered the maximum space available in the Petri dish. In tables 3, 4, 5, 6, 7, and 8, white mycelium and maximum growth are designated by 15, as derived from the

Table 9.—Color reactions of isolates of the gladiolus *Fusarium* on steamed rice. Colors are from the manual of Ridgway (1912).

ISOLATE	COLOR OF SUBSTRATUM	
	After 3 Weeks	After 11 Weeks
45-73.....	Thulite Pink	Pale Purplish Gray to Deep Purplish Gray
45-80.....	Dahlia Carmine	Bluish Black and Tawny Olive
46-3.....	White	Tawny Olive
46-5.....	Indian Lake	Dull Violet Black and Tawny Olive
46-9.....	Spinel Red	Dull Violet Black and Tawny Olive
46-14.....	Thulite Pink to Dull Dusky Purple	Dull Violet Black and Tawny Olive
47-6.....	Spinel Red	Dull Violet Black
47-10.....	Indian Lake	Bluish Black and Tawny Olive
49-4.....	Spinel Red to Indian Lake	Tawny Olive and Bluish Black
49-15.....	Indian Lake	Bluish Black to Tawny Olive
49-23.....	Indian Lake	Tawny Olive and Bluish Black
49-30.....	Indian Lake to Dull Dusky Purple	Dull Violet Black
49-31.....	Spinel Pink	Purplish Gray and Tawny Olive
50-6.....	Spinel Red to Indian Lake	Bluish Black
50-24.....	Salmon Buff, Thulite Pink, and Indian Lake	Purplish Gray and Tawny Olive
50-27.....	Thulite Pink to Dark Maroon Purple	Payne's Gray to Bluish Black
50-28.....	Indian Lake	Dull Violet Black
45-8.....	Spinel Red to Indian Lake	Dull Violet Black
45-74.....	Thulite Pink to Indian Lake	Tawny Olive to Dresden Brown
45-75.....	Indian Lake	Blackish Violet Gray and Bluish Slate Black
45-78.....	Indian Lake to Bordeaux	Dark Violet Gray
46-4.....	Spinel Pink to Spinel Red	Plumbago Slate and Old Gold
46-12.....	Dahlia Carmine	Dusky Slate Blue
47-1.....	Thulite Pink	Tawny Olive and Plumbago Slate
47-8.....	Light Buff	Clay Color to Saccardo's Amber
47-12.....	Thulite Pink	Deep Plumbago Gray and Tawny Olive
47-19.....	Thulite Pink to Indian Lake	Tawny Olive and Dusky Purplish Gray
47-32.....	Spinel Pink	Dull Violet Black
49-1.....	Thulite Pink to Indian Lake	Sudan Brown and Deep Purplish Gray
49-17.....	Indian Lake	Dull Purplish Black
49-19.....	Light Buff	Old Gold
50-7.....	Thulite Pink to Indian Lake	Dull Purplish Black and Dusky Slate Blue
50-22.....	Thulite Pink	Plumbago Slate and Old Gold
50-25.....	Dull Violet Black	Plumbago Slate
47-2.....	Spinel Pink to Indian Lake	Dull Violet Black and Dusky Slate Blue
47-3.....	Thulite Pink	Plumbago Slate and Old Gold
49-8.....	Indian Lake to Dull Violet Black	Dull Violet Black and Dusky Slate Blue
49-20.....	Cameo Pink	Raw Sienna
50-23.....	Indian Lake to Dull Violet Black	Dusky Slate Blue
50-26.....	Thulite Pink to Indian Lake	Plumbago Slate

Coons & Strong (1931) scheme shown on pages 458 and 459.

With the exception of isolates 50-26, 49-8, and 50-24, the isolates that were the most sensitive to the copper salts were also the most sensitive to mercuric chloride. Isolates from the three disease forms could not be separated on the basis of their reactions to mercuric chloride.

Although the isolates used in this study varied considerably in their sensitivity to mercuric chloride, as well as to other chemicals, more intensive work would have to be done before a conclusion could be reached that differences in sensitivity are responsible for differences in disease control obtained in tests involving chemical treatment of corms. However, sensitivity differences possibly could be factors contributing to control differences.

Color Reactions on Steamed Rice

Steamed rice was recommended by Wollenweber *et al.* (1925) as being especially useful in identification of groups of *Fusarium* because of the distinctive colors produced by cultures grown on it. Nelson, Coons, & Cochran (1937) reported that isolates from two forms of *Fusarium* yellows of celery fell into two groups when grown on steamed rice; in all instances the cultures which formed color on rice produced only form I of *Fusarium* yellows, while the cultures that were colorless produced only form II.

In the present study on the gladiolus *Fusarium*, 2 grams of rice and 6 milliliters of distilled water were placed in each of 80 test tubes. The tubes were plugged with cotton and steamed 1 hour on each of 3 successive days. Then a small amount of an agar slant culture was transferred to each tube of steamed rice. Two tubes of rice were used for each isolate. The test was run twice. In one trial the inoculated tubes were kept in the dark in an incubator held at 25 degrees C. In the other trial the tubes, in wire baskets, were placed on a cabinet shelf so that they were exposed to diffused light during the day. Colors produced in the rice substratum were, after 3 weeks and again after 11 weeks, compared with plates in the color manual of Ridgway (1912). The colors

produced in the two trials were almost identical.

As shown in table 9, the 40 isolates of gladiolus *Fusarium* varied a great deal in the colors they produced on steamed rice. As will be described more fully in a section on pH studies, isolates of *Fusarium* caused progressive pH changes to occur in the culture medium. In the first 3 weeks, when the cultures were acid in reaction, the colors in most cases were forms of pink or red, although one isolate remained white and several others were shades of violet or purple. In the last 6 or 7 weeks, when the cultures were alkaline in reaction, the colors changed to blues, violets, or purples, in many of which olives and browns appeared. There was no relation between the disease form from which the isolates were obtained and the colors the isolates produced on rice.

VARIATIONS IN CULTURE TYPES AND PATHOGENICITY

Variability in culture types and in pathogenicity within a given species of *Fusarium* seems to be of almost universal occurrence. The only exception found in a search through the literature was in the report by Blank (1934) that different strains of the cabbage yellows organism were uniform in their cultural behavior and pathogenicity.

The relation between growth types of cultures of *Fusarium* on laboratory media and differences in pathogenicity has been studied by many workers. Ullstrup (1935) reported that with the *Fusarium* stage of *Gibberella saubinetii* (Mont.) Sacc. rapid mycelial growth and abundant aerial mycelium are directly correlated with a high degree of pathogenicity in the majority of cases. Earlier, Brown (1928), working with certain fruit-rotting species of *Fusarium*, had pointed out that the mycelial type of culture is the most pathogenic. He stated that this type of growth is the form generally obtained in first isolations from diseased tissues. Harvey (1929) reported that high virulence in various strains of *Fusarium fructigenum* Fries was correlated with vigorous mycelial growth. Armstrong, MacLachlan, & Weindling (1940) reported that in the cotton-wilt organism,

Fusarium vasinfectum Atk., cultures that exhibited abundant aerial mycelium and grew rapidly were in the highly pathogenic group, but that variation from this cultural type may or may not be paralleled by decrease in pathogenicity. Wellman & Blaisdell (1941), in a study of pathogenic and cultural variation among single-spore isolates from strains of the tomato-wilt *Fusarium*, reported that the raised form is the most highly pathogenic, the appressed is mildest, and intermediate cultural types are intermediate in pathogenicity. In a later study, Wellman (1942) reported that these pathogenically and culturally variable strains of the tomato-wilt *Fusarium* also could be distinguished by differences in pH relations.

Many studies have been made on changes occurring in laboratory cultures of *Fusarium*. Burkholder (1925) reported that an isolate of *Fusarium martii phaseoli* Burkh. lost some of its virulence during its 5 years in culture. He noted changes in morphology and physiology, also. When first isolated, the pathogen produced on most media a blue-green slimy growth. At the end of 6 years the growth was white and fluffy. Armstrong, MacLachlan, & Weindling (1940) reported that variations of the cotton-wilt *Fusarium* were chiefly in two directions: decrease in abundance of aerial mycelium and decrease in the rate of radial growth. No changes occurred in the opposite direction. Isolates that had long been retained in culture were weakly pathogenic. Their cultural characteristics indicated that they were variants which had arisen in culture. These same authors reported that a cultural variant may or may not be less pathogenic than the isolate from which it has arisen.

McCulloch (1944) noted that variations and changes in pathogenicity occurred in isolates of the organism she called *Fusarium orthoceras* App. et Wr. var. *gladioli*. She observed that pathogenicity of this vascular *Fusarium* was reduced by long periods of culture in artificial media but she kept no extensive records of the changes. She also found that the isolates varied in their ability to infect different gladiolus varieties. Although she observed that some isolates produced dense

aerial mycelium and others were of the appressed type, she made no attempt to associate these characters with differences in pathogenicity.

Several investigators have reported cases of physiologic specialization in certain species of *Fusarium*. Broadfoot (1926) reported that at least nine physiologic forms of *F. lini* Bolley can be distinguished by their parasitism on four varieties of flax. Armstrong & Armstrong (1950) wrote that there are definitely two

Table 10.—Growth forms of isolates of the gladiolus *Fusarium* when originally cultured on potato dextrose agar and when grown on Wellman's agar, August, 1953.

ISOLATE	GROWTH FORM OF ORIGINAL ISOLATES ON POTATO DEXTROSE AGAR	GROWTH FORM ON WELLMAN'S AGAR, 1953
45-73...	Not recorded	Raised
45-80...	Not recorded	Intermediate-appressed
46-3...	Not recorded	Appressed
46-5...	Intermediate	Intermediate-appressed
46-9...	Intermediate	Intermediate-raised
46-14...	Not recorded	Intermediate-raised
47-6...	Raised	Intermediate-raised
47-10...	Raised	Intermediate-raised
49-4...	Intermediate	Raised
49-15...	Raised	Appressed
49-23...	Raised	Raised
49-30...	Raised	Intermediate-raised
49-31...	Intermediate	Appressed
50-6...	Not recorded	Intermediate-raised
50-24...	Not recorded	Intermediate-raised
50-27...	Not recorded	Raised
50-28...	Not recorded	Intermediate-raised
45-8...	Raised	Raised
45-74...	Not recorded	Appressed
45-75...	Not recorded	Intermediate-raised
45-78...	Not recorded	Intermediate-appressed
46-4...	Appressed	Intermediate-raised
46-12...	Intermediate	Intermediate-raised
47-1...	Appressed	Raised
47-8...	Appressed	Intermediate-appressed
47-12...	Not recorded	Raised
47-19...	Appressed	Intermediate-raised
47-32...	Raised	Raised
49-1...	Intermediate	Appressed
49-17...	Appressed	Appressed
49-19...	Intermediate	Intermediate-raised
50-7...	Not recorded	Intermediate-appressed
50-22...	Not recorded	Intermediate-raised
50-25...	Not recorded	Intermediate-raised
47-2...	Appressed	Intermediate-raised
47-3...	Appressed	Raised
49-8...	Raised	Intermediate-raised
49-20...	Intermediate	Intermediate-appressed
50-23...	Not recorded	Raised
50-26...	Not recorded	Raised

and probably more biological races of *F. oxysporum* f. *tracheiphilum* Sny. & Hans. on the basis of pathogenicity on varieties of soybeans and cowpeas.

Studies on the relation of culture types and pathogenicity of the 40 isolates of gladiolus *Fusarium* listed in table 1 were undertaken by the writer with the hope of finding a possible relation between colony type, disease form, and degree of virulence. Methods of determining virulence are described under "Pathogenicity Tests."

Culture Types of the Gladiolus *Fusarium*.—Original isolations from diseased gladiolus corms were made on potato dextrose agar. Growth forms of the

original cultures were not recorded for all isolates. However, 23 of the isolates were classified as belonging to the raised, intermediate, or appressed types on the basis of their aerial mycelium, table 10. Later, all isolates were grown on the medium found by Wellman (1942) to give more distinctive reactions between cultural variants. This medium, referred to as Wellman's agar in the present investigation, has the following composition: proteose peptone 5.0 grams, dihydrogen potassium phosphate 0.5 gram, magnesium sulfate 0.5 gram, maltose 15.0 grams, ferrous sulfate 0.03 gram, agar 12.0 grams, water 1,000.0 milliliters.

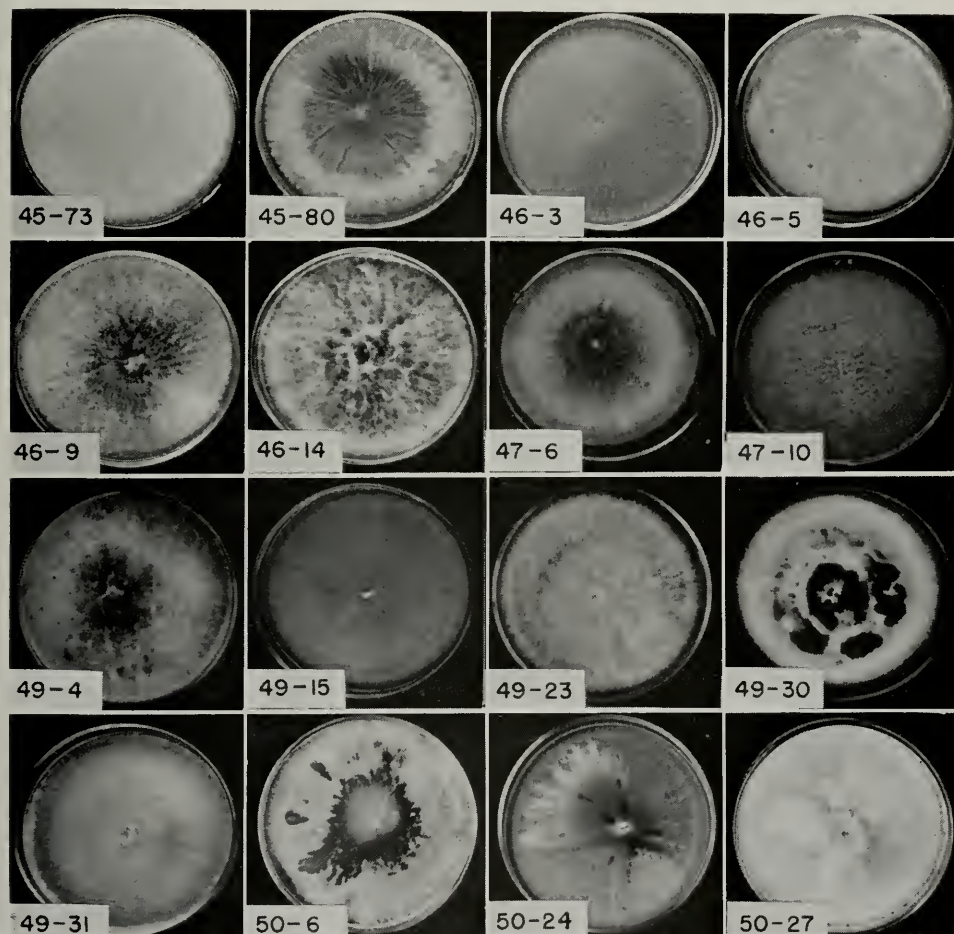


Fig. 13.—Raised, appressed, and intermediate growth forms of vascular isolates of *Fusarium* grown on Wellman's agar. The extreme raised and extreme appressed forms are shown in cultures 45-73 and 49-15, respectively. Cultures are 20 days old.

Growth forms, based on the classification of Wellman & Blaisdell (1941), of the 40 isolates of the gladiolus *Fusarium* as these isolates appeared in August, 1953, are given in table 10. When cultures of like form were grouped together, 11 were classified as raised, 17 as

intermediate-raised, 6 as intermediate-appressed, and 6 as appressed. Photographs of 36 of the cultures are shown in figs. 13 and 14.

Table 10 shows that by 1953 some of the cultures were no longer of the same type as the original isolations. When these

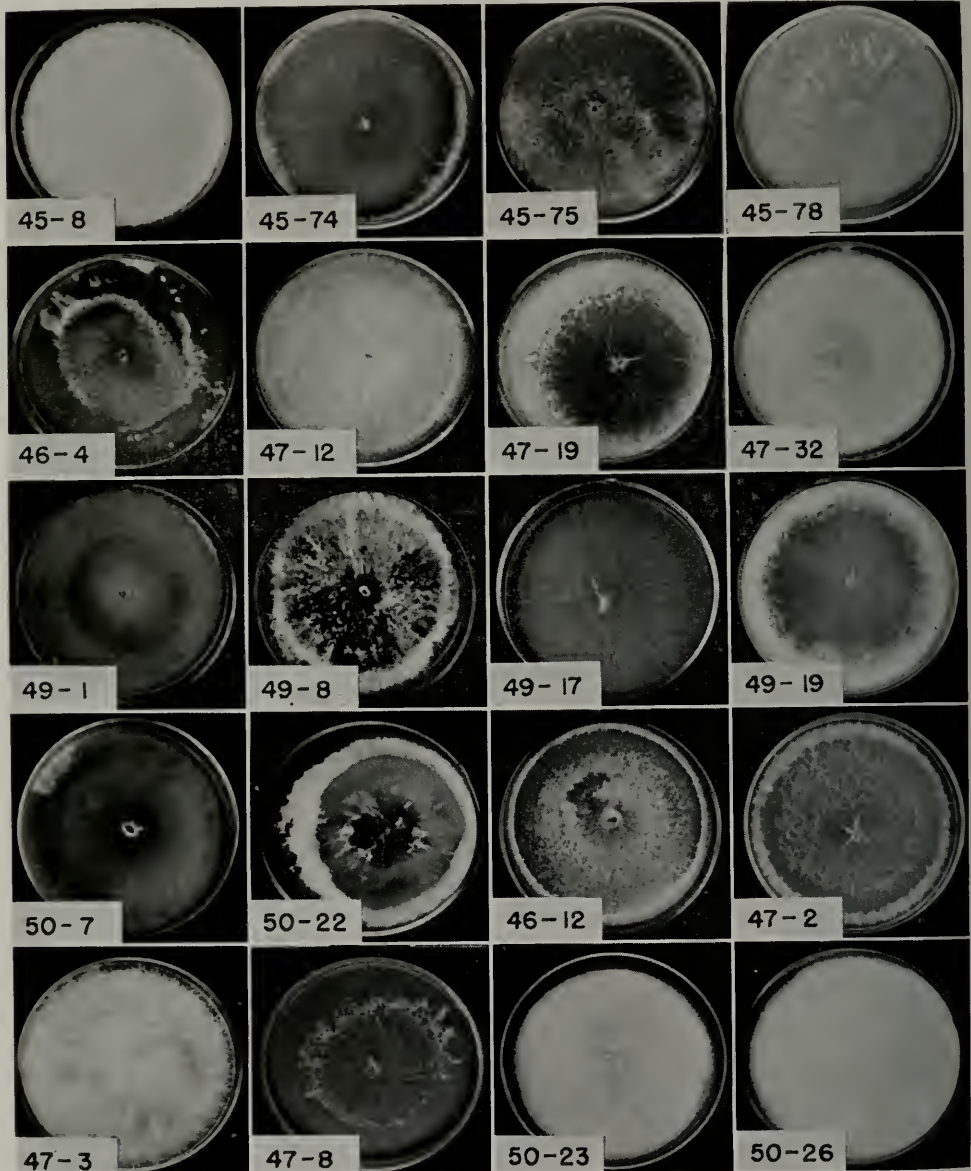


Fig. 14.—Raised, appressed, and intermediate growth forms of brown rot and basal dry rot isolates of *Fusarium* grown on Wellman's agar. Cultures 45-8 through 50-22 in the upper four rows are brown rot isolates; cultures 46-12 through 50-26 in the two bottom rows are basal dry rot isolates. Cultures are 20 days old.

Table 11.—Changes in growth form of isolates of the gladiolus *Fusarium* after being grown for various periods of time on laboratory media.

DIRECTION OF CHANGE	NUMBER OF ISOLATES
No change.....	9
Raised to intermediate....	4
Raised to appressed.....	1
Intermediate to raised....	1
Intermediate to appressed..	2
Appressed to intermediate..	4
Appressed to raised.....	2

changes were classified, with the two intermediate forms included in a single group, the results shown in table 11 were obtained.

Records show that variation in these cultures took place in both directions. Of the 14 isolates in which change in culture type was observed, 7 changed from raised to or toward appressed and 7 changed in the opposite direction.

pH Studies.—Wellman (1942) reported that raised (virulent) and appressed (mild) strains of the tomato-wilt *Fusarium* produced characteristic progressive pH changes in a liquid culture medium. Immediately after either of the strains began growing, acidity increased in the liquids for the first 5 days and became most intense in cultures of the appressed organism. The pH readings then gradually rose, but not at the same rate, until a maximum pH of about 8.7 was reached. With the raised strains, neutrality was reached at about the twelfth day, but with the appressed strains it was not reached until about the twenty-fifth day. By this time the pH of raised strains had nearly reached the maximum. The appressed strains did not reach the same point until after the fortieth day.

The 40 isolates of gladiolus *Fusarium* cultured in the studies reported here were grown in Tochinai liquid of the following composition: proteose peptone 10.0 grams, dihydrogen potassium phosphate 0.50 gram, magnesium sulfate 0.25 gram, maltose 20.0 grams, water 1,000.0 milliliters. The original pH of the medium was 6.21. Each isolate was grown in 100 milliliters of the culture medium in a 250-milliliter Erlenmeyer flask. Uniform disks of inoculum

were cut from agar plate cultures with a sterile metal tube 9 millimeters in diameter. Each flask was inoculated with a single disk of inoculum. The flasks were shaken immediately and then left on the laboratory table. Small samples of liquid were removed from the flasks for pH determination after 3, 5, 8, 12, 15, 23, 28, 34, 41, 50, and 58 days. All pH determinations were made with a Beckman pH meter, standardized at frequent intervals against known buffer solutions.

The pH readings for 38 isolates are shown in table 12. As isolates 49-15 and 50-27 became contaminated early in the experiment they are not included in the table.

The progressive changes in pH produced by 16 of the isolates are shown in figs. 15, 16, 17, and 18. Pairs of isolates which had shown apparent differences in pathogenicity were selected from each disease form for comparison in pH reactions. The more virulent isolate of each pair, as determined by inoculation tests, is represented by the solid line in the graphs, and the milder isolate is represented by the broken line.

The figures show curves of the same general type as that obtained by Wellman for the tomato *Fusarium*. The liquid medium became more acid through the period of the first 5 days; then it gradually became less acid and more alkaline until a maximum of about pH 8.5 was reached.

In fig. 15A, vascular isolates 49-4 and 49-23 are compared. Both isolates were of the raised type on Wellman's agar, fig. 13. Isolate 49-4 was much more virulent than isolate 49-23 in pathogenicity tests in the greenhouse. Also, isolate 49-4 caused rot on corms inoculated in the laboratory; isolate 49-23 did not. The progressive pH changes produced by these two isolates were quite similar.

In fig. 15B, vascular isolates 50-24 and 50-28 are compared. Both isolates were of the intermediate-raised type on Wellman's agar. In pathogenicity tests, isolate 50-28 was the more virulent under both greenhouse and laboratory conditions. Isolate 50-28 reached a lower pH reading than isolate 50-24 and also required a longer time to reach maximum alkalinity. In these two isolates the relation between

Table 12.—Readings in pH of Tochinai liquid inoculated with different isolates of the *gladiolus Fusarium*; original pH 6.21.

ISOLATE	DAYS AFTER INOCULATION										
	3	5	8	12	15	23	28	34	41	50	58
45-73	5.68	5.25	4.87	4.84	4.97	6.99	7.45	8.36	8.48	8.47	8.32
45-80	6.10	4.78	4.94	5.52	6.06	7.10	7.68	8.27	8.52	8.52	8.47
46-3	5.63	5.10	4.93	5.30	6.03	6.50	7.38	8.29	8.54	8.58	8.45
46-5	5.51	4.85	4.73	5.60	6.65	8.49	8.72	8.72	8.55	8.15	8.59
46-9	5.50	4.84	4.75	5.49	6.06	6.85	8.22	8.63	8.61	8.48	8.41
46-14	5.55	4.83	4.78	5.78	6.03	6.57	7.90	8.40	8.59	8.54	8.45
47-6	5.60	4.78	4.63	5.52	5.84	8.53	8.50	8.61	8.48	8.32	8.12
47-10	5.33	4.78	4.63	5.73	6.68	6.77	6.47	8.21	8.44	8.54	8.61
49-4	5.55	4.90	4.95	6.20	6.47	6.32	7.53	8.44	8.62	8.42	8.45
49-23	5.78	4.92	4.90	6.48	7.03	6.25	7.07	8.31	8.46	8.50	8.38
49-30	5.94	5.30	4.88	5.79	6.88	7.93	8.40	8.58	8.52	8.32	8.15
49-31	5.27	5.03	4.83	4.98	6.00	7.50	8.02	8.53	8.51	8.44	8.43
50-6	5.59	4.85	4.89	6.08	6.42	6.68	6.98	8.45	8.48	8.65	8.62
50-24	5.41	4.92	4.95	6.38	6.98	8.37	8.50	8.50	8.40	8.23	8.09
50-28	5.55	4.75	4.61	5.38	6.07	6.53	6.71	7.82	8.45	8.55	8.53
45-8	5.60	5.00	4.88	5.89	7.38	8.49	8.55	8.45	8.47	8.32	8.15
45-74	5.70	5.03	4.92	5.95	6.67	6.63	7.58	8.37	8.43	8.61	8.50
45-75	5.45	4.88	4.91	6.18	7.00	6.98	7.32	8.19	8.44	8.51	8.40
45-78	5.88	5.21	4.78	5.61	6.70	8.34	8.37	8.48	8.58	8.55	8.38
46-4	5.75	5.45	5.27	5.58	6.26	7.33	7.33	8.40	8.48	8.64	8.51
46-12	5.51	4.74	4.60	5.12	5.50	5.93	6.17	8.20	8.42	8.58	8.45
47-1	5.69	5.05	4.93	5.55	6.38	6.95	7.84	8.48	8.59	8.50	8.39
47-8	5.59	5.27	5.04	5.91	6.38	6.77	7.61	8.37	8.57	8.65	8.45
47-12	5.64	5.18	4.94	4.90	5.10	6.55	7.59	7.48	8.43	8.58	8.45
47-19	5.68	5.12	5.00	4.96	5.24	6.60	7.54	8.08	8.62	8.49	8.42
47-32	5.69	5.12	4.86	5.16	6.15	8.28	8.40	8.62	8.48	8.44	8.28
49-1	5.83	5.10	4.89	5.75	6.70	7.19	7.35	8.48	8.61	8.59	8.41
49-17	5.42	4.89	4.77	6.00	6.67	7.29	7.44	8.29	8.54	8.54	8.46
49-19	5.53	4.95	4.89	6.40	7.10	6.47	8.15	8.40	8.52	8.62	8.47
50-7	5.50	5.21	5.07	5.96	6.17	6.70	7.27	8.40	8.64	8.49	8.46
50-22	5.66	4.95	4.78	5.74	6.73	7.48	8.06	8.44	8.43	8.50	8.52
50-25	5.48	5.09	4.95	6.50	6.85	7.51	7.69	8.29	8.60	8.65	8.49
47-2	5.72	4.87	4.55	5.02	5.81	7.15	8.02	8.48	8.37	8.60	8.48
47-3	5.51	4.90	4.88	6.15	7.08	7.94	8.39	8.45	8.55	8.58	8.42
49-8	5.72	4.90	4.62	5.67	6.38	6.32	6.10	8.23	8.41	8.47	8.51
49-20	5.53	4.93	4.80	6.13	6.64	6.52	7.38	8.32	8.43	8.65	8.59
50-23	5.52	4.85	4.71	6.04	6.60	7.34	6.50	8.36	8.39	8.56	8.43
50-26	5.67	5.20	4.85	4.83	5.68	8.00	8.48	8.48	8.38	8.33	8.23

pH changes and virulence is exactly opposite to that reported by Wellman for the tomato *Fusarium*.

In fig. 16A, brown rot isolates 47-12 and 50-25 are compared. Isolate 47-12 was of the raised type on Wellman's agar; 50-25 was intermediate-raised. Isolate 47-12 produced rot in inoculated corms; 50-25 did not. The pH curves of the two isolates show that in cultures of both isolates the same low point was reached at about the same time but that the rise in pH was much sharper in cultures of isolate 50-25 than it was in cultures of isolate 47-12. With isolate 50-25, neutrality was reached in about 17 days but, with isolate 47-12, not until after 25 days.

Brown rot isolates 49-17 and 49-19 are compared in fig. 16B. Isolate 49-17 was of the appressed type, and isolate 49-19 was intermediate-raised. Isolate 49-17 produced corm rot in laboratory tests in 1952 and 1953; isolate 49-19 produced corm rot in 1952 but not in 1953. The pH curves for these two isolates are similar except for a second drop in pH from the fifteenth to the twenty-third days in the culture of isolate 49-19.

Vascular isolates 45-73 and 46-3 are compared in fig. 17A. Isolate 45-73 was the more virulent in greenhouse tests in 1948 and 1953. In 1952, isolate 46-3 was slightly more virulent. Isolate 45-73 was of the raised type; 46-3 was appressed.

The pH curves for these two isolates are quite similar. Isolate 45-73 remained in the low acid range a little longer but

reached the alkaline range ahead of isolate 46-3. After 30 days, the pH curves of these two isolates were nearly identical.

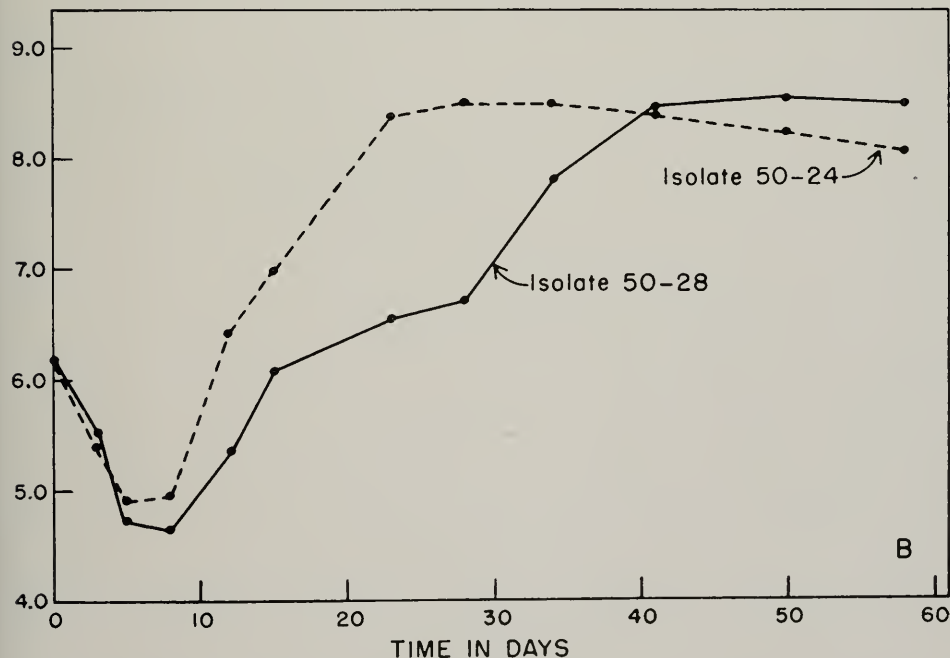
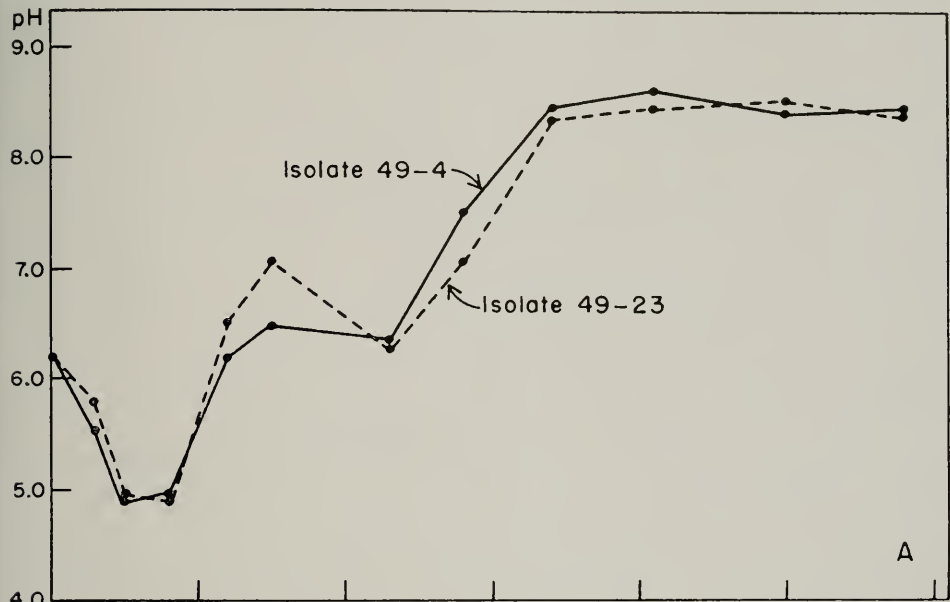


Fig. 15.—Progressive changes in pH produced in Tochinai liquid by four vascular isolates of *Fusarium*.

In fig. 17B, brown rot isolates 50-7 50-22 was intermediate-raised. Isolate 50-7 50-22 was the more virulent in laboratory and greenhouse tests. The pH curves for

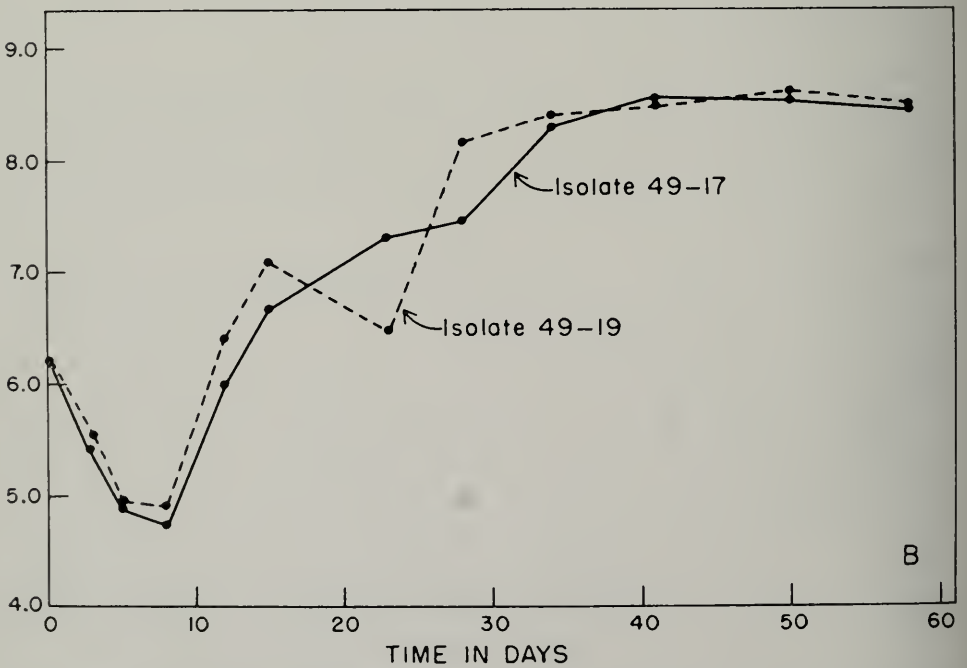
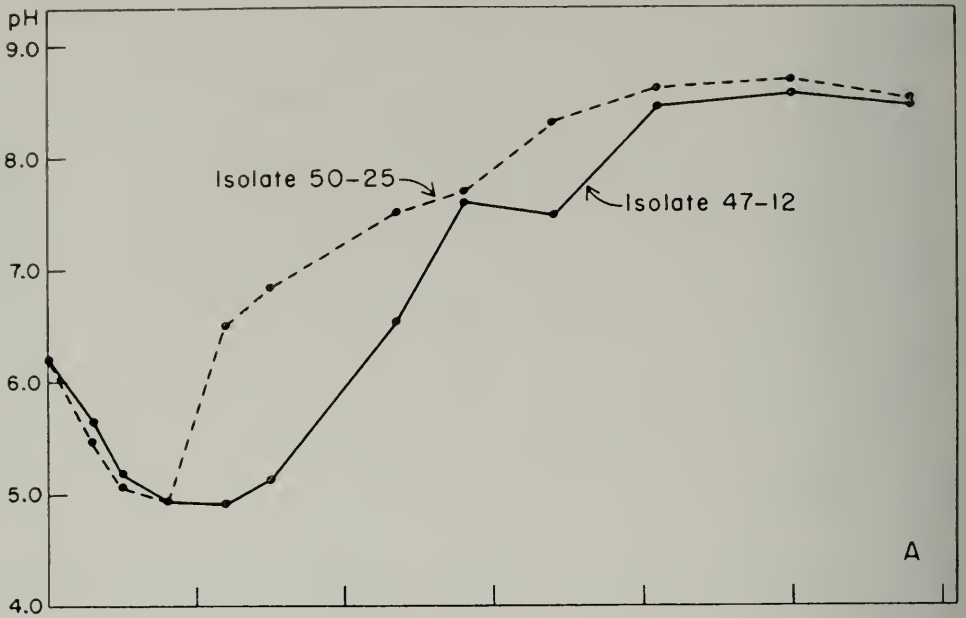


Fig. 16.—Progressive changes in pH produced in Tochnai liquid by four brown rot isolates of *Fusarium*.

these two isolates are similar except for a divergence that occurred between the twelfth and thirty-fourth days.

Two sets of basal dry rot isolates are compared in fig. 18. In fig. 18*A*, isolate 47-2 was of the intermediate-raised type

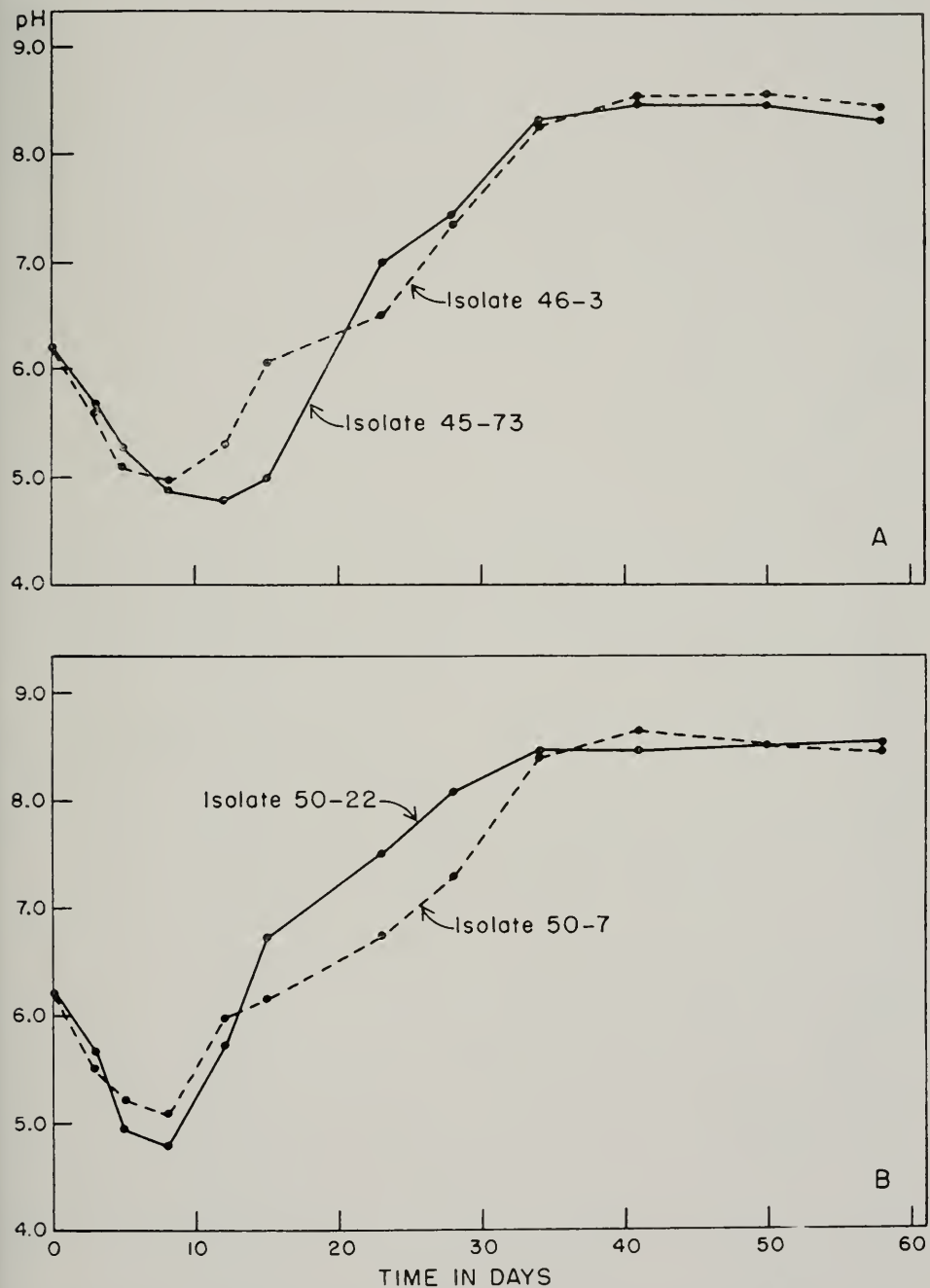


Fig. 17.—Progressive changes in pH produced in Tochinai liquid by, *A*, two vascular isolates and, *B*, two brown rot isolates of *Fusarium*.

and 47-3 was raised. Isolate 47-3 was the more virulent in greenhouse tests; 47-2 was slightly more virulent than 47-3 in

the laboratory. Isolate 47-2 produced the greater amount of acid during early growth and its pH readings remained

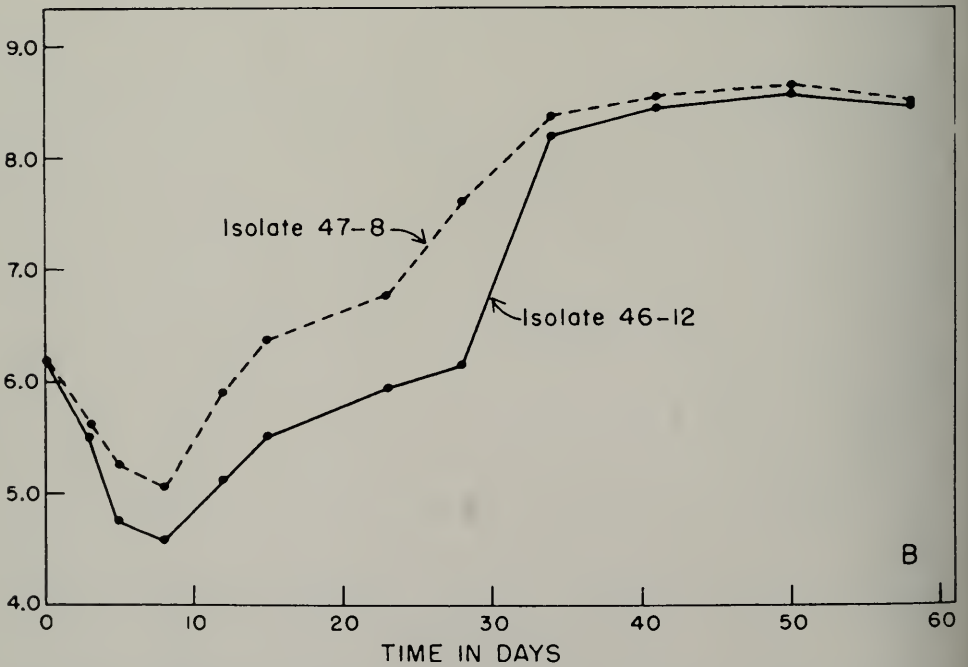
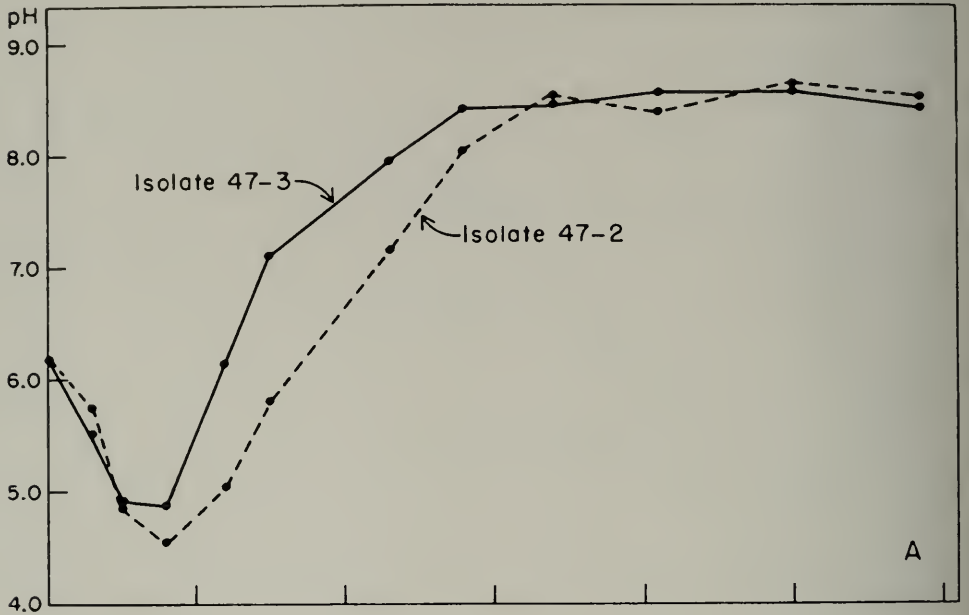


Fig. 18.—Progressive changes in pH produced in Tochinai liquid by four basal dry rot isolates of *Fusarium*.

lower than those for isolate 47-3 until the thirty-fourth day.

In fig. 18B, isolate 46-12 was intermediate-raised, and isolate 47-8 was intermediate-appressed. Isolate 46-12 was more virulent in laboratory tests. In this case the pH readings for the more virulent isolate were lower than those for the milder isolate from the beginning until the fifty-eighth day.

In these experiments no consistent relations between culture types, pH changes, or degrees of virulence were observed. In this respect, the gladiolus *Fusarium* appears to differ from the tomato *Fusarium*.

MORPHOLOGY

A complete morphological study of all the isolates from the vascular, brown rot, and basal dry rot forms of the gladiolus *Fusarium* disease was not attempted. Some of the isolates produced the typically 2- to 5-septate macrospores rather consistently, but many isolates produced only the single-celled type. In some isolates, septate spores were very abundant in the original cultures from diseased gladiolus corms, but, after one or two transfers, only single-celled spores could be found. Spores seemed to be produced more abundantly on Well-

Table 13.—Measurements of conidia produced by six isolates of the gladiolus *Fusarium* on Wellman's agar.

DISEASE FORM	ISOLATE AND TYPE OF CONIDIA	MEAN, MICRONS	RANGE, MICRONS
Vascular	Isolate 49-15		
	0-septate	9.2 x 3.3	5.5-13.1 x 2.8-4.1
	1-septate	16.0 x 4.0	6.9-27.6 x 2.8-5.5
	2-septate	26.2 x 4.1	16.6-34.5 x 3.4-4.8
	3-septate	32.1 x 4.1	23.5-45.5 x 3.4-4.1
	4-septate	45.3 x 3.9	37.3-56.6 x 2.8-4.1
Vascular	5-septate	53.8 x 4.1	Only 1 spore
	Isolate 50-27		
	0-septate	8.8 x 3.0	5.5-12.4 x 2.8-4.1
	1-septate	16.7 x 3.5	11.7-23.5 x 2.8-4.8
	2-septate	23.2 x 4.0	19.3-37.3 x 2.8-5.5
	3-septate	35.1 x 4.2	24.8-44.2 x 4.1-5.5
Brown rot	4-septate	42.7 x 4.1	34.5-48.3 x 4.1-4.1
	5-septate	46.4 x 4.5	44.2-53.8 x 4.1-5.5
	Isolate 45-74		
	0-septate	9.9 x 3.3	6.9-15.2 x 2.8-4.1
	1-septate	18.7 x 3.9	9.7-24.8 x 2.8-4.1
	2-septate	24.5 x 4.0	17.9-31.7 x 2.8-4.8
Brown rot	3-septate	30.2 x 4.1	15.2-40.0 x 2.8-5.5
	4-septate	34.5 x 5.2	34.5-34.5 x 4.8-5.5
	5-septate	None	—
	Isolate 50-25		
	0-septate	8.9 x 3.0	5.5-13.8 x 2.1-4.1
	1-septate	15.2 x 3.8	11.0-19.3 x 2.8-4.8
Basal dry rot	2-septate	19.7 x 4.0	15.2-24.8 x 2.8-5.5
	3-septate	31.7 x 4.3	20.7-44.2 x 4.1-5.5
	4-septate	39.7 x 4.3	37.3-44.2 x 4.1-5.5
	5-septate	44.2 x 4.1	Only 1 spore
	Isolate 49-20		
	0-septate	9.1 x 3.4	6.9-15.2 x 2.8-4.8
Basal dry rot	1-septate	16.3 x 4.0	9.7-29.0 x 2.8-4.1
	2-septate	22.3 x 4.2	13.8-31.7 x 3.4-5.5
	3-septate	29.4 x 4.4	22.1-37.3 x 4.1-5.5
	4-septate	29.0 x 4.1	Only 1 spore
	5-septate	None	—
	Isolate 50-26		
0-septate	9.6 x 4.0	4.1-22.1 x 2.8-5.5	
Basal dry rot	1-septate	17.6 x 4.2	12.4-23.5 x 3.4-4.8
	2-septate	22.9 x 4.4	17.9-29.0 x 3.4-5.5
	3-septate	31.8 x 5.2	24.8-40.0 x 4.1-5.5
	4-septate	32.4 x 5.2	31.7-33.1 x 4.8-5.5
	5-septate	None	—

man's agar than on potato dextrose agar.

From several isolates which produced septate macrospores rather abundantly on Wellman's agar, two isolates from each

disease form were selected for spore measurements. The only purpose of making spore measurements was to find out if isolates from the different disease forms

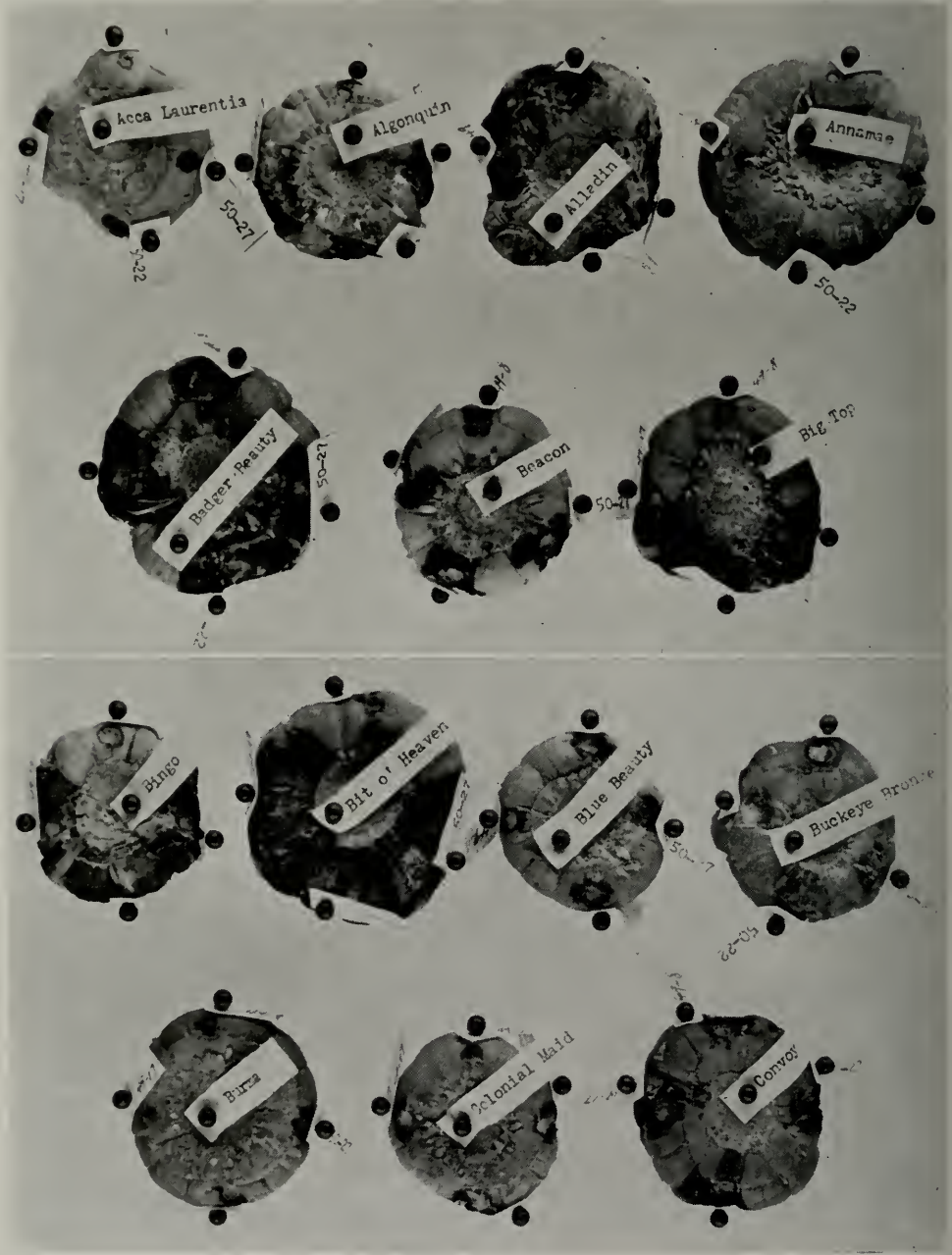


Fig. 19.—Corns of 14 gladiolus varieties, each corm inoculated with four isolates of *Fusarium*. The locations of the inoculations on each corm are as follows: top, isolate 49-8; right, 50-27; bottom, 50-22; left, 49-17. Photograph was taken 34 days after inoculation.

might be distinguished by differences in spore sizes. As pointed out by Harter (1939), several factors influence the size of *Fusarium* spores produced in laboratory culture. It is often difficult, if not impossible, by any means of manipulation of the culture to obtain a morphological agreement with the description of a given species. It seemed reasonable to assume, however, that a fair comparison could be made of spores from cultures which had been kept under the same conditions.

Six isolates were selected for use in making spore measurements and were grown on Wellman's agar in Petri dishes for 14 days in a location where the dishes were exposed to diffused light during daylight hours. All measurements were made within 2 or 3 days after the cultures were 14 days old. Cultures from which measurements could not be made immediately were placed in a refrigerator at 3-5 degrees C. until measurements could be made. The spores to be measured were mounted on 2 per cent plain agar on a microscope slide. Thirty spores of each septation type were measured from each culture in which this number of spores was available. The 4- and 5-septate types were so rare in most of the cultures that it was impossible to find 30 spores for measurement.

Mean measurements and ranges of measurements of spores from the six selected isolates are shown in table 13. The differences in spore sizes are not great enough to place the isolates in distinct groups on the basis of spore size.

PATHOGENICITY TESTS

In this investigation, the pathogenicity of the isolates of *Fusarium* was tested both in the laboratory and in the greenhouse. All 40 isolates were tested in the laboratory; 27 of them were tested in the greenhouse.

Laboratory Tests

Large mature gladiolus corms, free from blemishes, were selected for the laboratory inoculation tests. After the husks had been removed, the corms were washed well with water and then left on a table

until dry. Four small wounds, approximately equal distances apart, were made on the basal area of each corm with a three-sixteenths-inch metal drill bit turned rapidly with thumb and index finger. Uniform disks of inoculum were cut from agar plate cultures with a sterile metal tube three-sixteenths inch in diameter. One disk of inoculum was pressed gently into each wound. Each corm was inoculated with four isolates; each isolate was used on four corms in each of the various tests.

Inoculations were identified by paper tags pinned near the points of inoculation, fig. 19. Immediately after they had been inoculated, the corms were placed in moist chambers and left for 48 hours. They were then removed and placed on a table in the laboratory. The corms were examined frequently, and at the end of 4 to 6 weeks final disease readings were made.

These inoculations resulted in the development of three general types of lesions: severe, in which the rot progressed steadily from the point of inoculation until most of the corm was rotted, shown in the Bit o' Heaven corm in fig. 19; mild, in which the rot progressed very slowly and only a narrow brown band appeared around the point of inoculation, shown in the Acca Laurentia corm in fig. 19; healed, in which no rot developed and the inoculation wound corked over, shown in the Annamae corm in fig. 19.

A basis on which to compare virulence of the isolates of *Fusarium*, as well as to compare susceptibilities of gladiolus varieties, was derived from a modification of McKinney's (1923) formula for disease evaluation. Class values of 0, 1, and 2, respectively, were assigned to the healed, mild, and severe types of lesions. Indexes of rot severity were calculated by use of the following formula:

$$\text{Rot severity index} = \frac{N_1 0 + N_2 1 + N_3 2}{2t} \times 100$$

where N_1, N_2, N_3 = number of corms in disease classes 1, 2, and 3, respectively

0, 1, 2 = values assigned to disease classes 1, 2, and 3 respectively

t = total number of corms used

Table 14.—Severity indexes for the rots produced by four isolates of *Fusarium* on 68 gladiolus varieties in 1951. The indexes were derived from the formula on page 481.

VARIETY	ISOLATE				AVERAGE
	49-8	49-17	50-22	50-27	
Abu Hassan.....	100.0	100.0	100.0	100.0	100.0
Acca Laurentia.....	37.5	50.0	50.0	25.0	40.6
Aladdin.....	75.0	100.0	100.0	87.5	90.6
Algonquin.....	50.0	100.0	100.0	100.0	87.5
Annamae.....	25.0	25.0	25.0	37.5	28.1
Badger Beauty.....	100.0	100.0	100.0	100.0	100.0
Beacon.....	62.5	87.5	100.0	100.0	87.5
Big Top.....	75.0	100.0	100.0	100.0	93.7
Bingo.....	75.0	100.0	100.0	100.0	93.7
Bit o' Heaven.....	62.5	100.0	100.0	100.0	90.6
Black Opal.....	87.5	75.0	100.0	100.0	90.6
Blue Beauty.....	62.5	50.0	87.5	62.5	65.6
Buckeye Bronze.....	37.5	75.0	87.5	87.5	71.9
Burma.....	50.0	12.5	50.0	50.0	40.6
Colonial Maid.....	37.5	75.0	100.0	100.0	78.1
Convoy.....	75.0	75.0	87.5	62.5	75.0
Corona.....	100.0	100.0	100.0	100.0	100.0
Crinkle Cream.....	87.5	100.0	100.0	100.0	96.9
Dieppe.....	50.0	100.0	100.0	100.0	87.5
Dr. F. E. Bennett.....	100.0	100.0	100.0	100.0	100.0
Dr. Whiteley.....	50.0	50.0	62.5	50.0	53.1
Dusty Miller.....	100.0	62.5	100.0	100.0	90.6
Early Rose.....	25.0	50.0	75.0	37.5	46.9
Elizabeth the Queen.....	75.0	100.0	100.0	100.0	93.7
Ethel Cave Cole.....	50.0	75.0	100.0	87.5	78.1
Fair Angel.....	50.0	75.0	87.5	50.0	65.6
Genghis Khan.....	100.0	100.0	100.0	100.0	100.0
Gloaming.....	62.5	100.0	100.0	100.0	90.6
Gold Eagle.....	50.0	37.5	62.5	62.5	53.1
Golden Dream.....	100.0	100.0	100.0	100.0	100.0
High Finance.....	62.5	100.0	100.0	100.0	90.6
Johan van Konynenburg.....	100.0	100.0	100.0	100.0	100.0
King Lear.....	75.0	100.0	100.0	100.0	93.7
King William.....	50.0	100.0	100.0	100.0	87.5
Lady Jane.....	12.5	75.0	87.5	87.5	65.6
Lantana.....	50.0	100.0	100.0	100.0	87.5
Larime.....	50.0	100.0	100.0	100.0	87.5
Leading Lady.....	50.0	100.0	100.0	87.5	84.4
Legend.....	50.0	75.0	87.5	50.0	65.6
Majuba.....	50.0	62.5	100.0	50.0	65.6
Malta.....	62.5	100.0	100.0	100.0	90.6
Margaret Beaton.....	50.0	50.0	50.0	50.0	50.0
Marguerite.....	62.5	75.0	100.0	100.0	84.4
Minuet.....	62.5	37.5	100.0	100.0	75.0
Miss Bloomington.....	50.0	50.0	50.0	50.0	50.0
Mother Kadel.....	50.0	100.0	100.0	100.0	87.5
Mrs. Lulu Hunt.....	50.0	50.0	50.0	50.0	50.0
New Europe.....	100.0	100.0	100.0	100.0	100.0
Ogarita.....	50.0	75.0	87.5	87.5	75.0
Ohio Nonpareil.....	50.0	87.5	100.0	87.5	81.2
Oklahoma.....	50.0	75.0	87.5	75.0	71.9
Oregon Gold.....	75.0	75.0	100.0	87.5	84.4
Pandora.....	75.0	100.0	100.0	100.0	93.7
Phyllis McQuiston.....	50.0	50.0	50.0	50.0	50.0
Prairie Gold.....	50.0	100.0	100.0	100.0	87.5
Purple Supreme.....	100.0	100.0	100.0	100.0	100.0
Red Charm.....	75.0	100.0	100.0	100.0	93.7
Rewi Fallu.....	75.0	100.0	100.0	100.0	93.7
Rosa Van Lima.....	25.0	75.0	62.5	37.5	50.0
Rose Ruffles.....	62.5	75.0	100.0	87.5	81.2

Table 14.—Concluded

VARIETY	ISOLATE				AVERAGE
	49-8	49-17	50-22	50-27	
Silentium.....	50.0	87.5	87.5	75.0	75.0
Silver Wings.....	75.0	100.0	100.0	100.0	93.7
Snow Princess.....	50.0	62.5	100.0	87.5	75.0
Southern Melody.....	100.0	100.0	100.0	100.0	100.0
Spirit of St. Louis.....	87.5	12.5	12.5	12.5	31.2
Valeria.....	37.5	37.5	87.5	50.0	53.1
Variation.....	100.0	100.0	100.0	100.0	100.0
Vulcan.....	50.0	75.0	100.0	87.5	78.1

Table 15.—Severity indexes for the rots produced by 40 isolates of *Fusarium* on seven gladiolus varieties in 1952. The indexes were derived from the formula on page 481.

ISOLATE	GLADIOLUS VARIETY							AVERAGE INDEX
	Ethel Cave Cole	Maid of Orleans	Miss Bloomington	Picardy	Rosa Van Lima	Snow Princess	Spirit of St. Louis	
45-73.....	0.0	12.5	0.0	37.5	0.0	0.0	25.0	10.7
45-80.....	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
46-3.....	0.0	0.0	0.0	25.0	0.0	0.0	0.0	3.6
46-5.....	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
46-9.....	0.0	0.0	0.0	12.5	0.0	0.0	12.5	3.6
46-14.....	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
47-6.....	0.0	0.0	0.0	50.0	0.0	0.0	0.0	7.1
47-10.....	0.0	0.0	0.0	25.0	0.0	0.0	0.0	3.6
49-4.....	12.5	0.0	0.0	100.0	0.0	62.5	0.0	25.0
49-15.....	0.0	0.0	12.5	0.0	0.0	0.0	0.0	1.8
49-23.....	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
49-30.....	0.0	0.0	0.0	100.0	0.0	0.0	0.0	14.3
49-31.....	0.0	0.0	0.0	25.0	0.0	0.0	0.0	3.8
50-6.....	0.0	0.0	12.5	75.0	0.0	0.0	0.0	12.5
50-24.....	0.0	0.0	0.0	25.0	0.0	0.0	0.0	3.8
50-27.....	0.0	0.0	0.0	75.0	0.0	0.0	0.0	10.7
50-28.....	0.0	12.5	12.5	75.0	0.0	0.0	0.0	14.3
45-8.....	0.0	0.0	0.0	62.5	0.0	0.0	0.0	8.9
45-74.....	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
45-75.....	12.5	0.0	0.0	75.0	0.0	0.0	0.0	12.5
45-78.....	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
46-4.....	12.5	0.0	0.0	25.0	12.5	0.0	0.0	7.1
46-12.....	37.5	0.0	25.0	100.0	0.0	0.0	0.0	23.2
47-1.....	87.5	0.0	37.5	87.5	12.5	0.0	0.0	19.6
47-8.....	0.0	0.0	12.5	37.5	0.0	0.0	0.0	7.1
47-12.....	0.0	0.0	0.0	100.0	0.0	0.0	0.0	14.3
47-19.....	0.0	0.0	12.5	100.0	0.0	0.0	0.0	16.1
47-32.....	0.0	0.0	25.0	100.0	0.0	0.0	0.0	17.7
49-1.....	0.0	0.0	0.0	100.0	0.0	0.0	0.0	14.3
49-17.....	25.0	12.5	37.5	100.0	0.0	37.5	0.0	30.3
49-19.....	0.0	0.0	50.0	100.0	0.0	37.5	0.0	26.8
50-7.....	25.0	0.0	50.0	100.0	0.0	37.5	37.5	35.7
50-22.....	100.0	12.5	62.5	100.0	0.0	100.0	12.5	55.3
50-25.....	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
47-2.....	37.5	37.5	62.5	87.5	50.0	12.5	87.5	53.6
47-3.....	62.5	12.5	50.0	100.0	25.0	12.5	12.5	39.3
49-8.....	12.5	0.0	12.5	87.5	0.0	0.0	75.0	26.8
49-20.....	0.0	0.0	50.0	87.5	0.0	0.0	25.0	23.2
50-23.....	12.5	0.0	25.0	100.0	0.0	0.0	12.5	21.4
50-26.....	0.0	0.0	25.0	100.0	0.0	0.0	0.0	17.8
Average.....	10.9	2.5	14.4	59.4	2.5	7.5	7.5	—

In 1951, four isolates were tested for ability to produce rot on corms of 68 varieties. Results are shown in table 14. Isolate 50-27 was a vascular isolate; the others were brown rot isolates. All four isolates caused rot on all varieties used in this test, but the severity of rot varied with isolates as well as with varieties. The average indexes in table 14 can be considered indicative of the relative susceptibility of the different varieties to *Fusarium* rot. Since the test was conducted under conditions that favored development of rot, varieties with an average index of 0 to 50 can be considered resistant and varieties with an index greater than 50 can be considered susceptible.

In 1952, all 40 isolates were tested on seven gladiolus varieties. The indexes of rot severity obtained in this test are shown in table 15. The isolates varied greatly in their ability to produce rot on corms of different varieties. Seven of the isolates failed to produce rot on any of the seven varieties tested. Only basal dry rot isolates 47-2 and 47-3 produced rot on all

seven varieties. The average rot severity index for the 7 basal dry rot isolates was 26.5, for the 16 brown rot isolates it was 17.8, and for the 17 vascular isolates it was 6.7. Apparently vascular isolates are less capable of causing corm rot than are the brown rot and basal dry rot isolates. These results are in general agreement with those of McCulloch (1944), who found that the vascular isolates with which she worked caused only mild rot on corms inoculated in the laboratory, while isolates from corms having the disease type described by Massey (1926) caused severe rot.

In this test the average severity index for rot on the variety Picardy was 59.4 while on Maid of Orleans and Rosa Van Lima it was only 2.5. The difference in susceptibility indicated by these figures agrees well with what is observed in commercial stocks of these varieties. *Fusarium* rot is very rare in stocks of Maid of Orleans and Rosa Van Lima but it is extremely common in stocks of Picardy.

Six of the varieties used in the 1951

Table 16.—Severity indexes for the rots produced in 2 successive years by four isolates of *Fusarium* on six gladiolus varieties.*

VARIETY AND YEAR	ISOLATE			
	49-8	49-17	50-22	50-27
Ethel Cave Cole				
1951.....	50.0	75.0	100.0	87.5
1952.....	12.5	25.0	100.0	0.0
Miss Bloomington				
1951.....	50.0	50.0	50.0	50.0
1952.....	12.5	37.5	62.5	0.0
Picardy†				
1951.....	75.0	100.0	100.0	100.0
1952.....	87.5	100.0	100.0	75.0
Rosa Van Lima				
1951.....	25.0	75.0	62.5	37.5
1952.....	0.0	0.0	0.0	0.0
Snow Princess				
1951.....	50.0	62.5	100.0	87.5
1952.....	0.0	37.5	100.0	0.0
Spirit of St. Louis				
1951.....	87.5	12.5	12.5	12.5
1952.....	75.0	0.0	12.5	0.0

*The indexes used in this table are taken from tables 14 and 15.

†In 1951, Silver Wings, a sport of Picardy, was used instead of Picardy.

test were used also in the 1952 test. The severity indexes for the rots caused by the same four isolates on corms of these varieties in the 2 years are shown in table 16. In 17 cases the rot indexes were lower in 1952 than they had been in 1951, in 5 cases they remained the same, and in 2 cases only they were higher.

A third test was made in 1953 when all 40 isolates were used on the varieties Picardy and Spirit of St. Louis. The results obtained with these varieties in 1952 and 1953 are shown in table 17. In the tests on Picardy the rot indexes in 27 isolates were lower in 1953 than in 1952, in 4

they were higher, and in 9 they remained the same. With Spirit of St. Louis the rot indexes in 5 isolates were lower in 1953 than in 1952, in 7 they were higher, and in 28 they remained the same. The indexes remaining unchanged included 7 instances in Picardy and 25 in Spirit of St. Louis in which no rot was produced in either year.

Although no attempt was made to maintain the same conditions for all tests, it is not likely that laboratory conditions varied greatly during the three tests. Differences in corm lots used in different years may have been partly responsible for

Table 17.—Severity indexes for the rots produced in 2 or 3 successive years by 40 isolates of *Fusarium* on two gladiolus varieties. Indexes were derived from the formula on page 481.

ISOLATE	PICARDY			SPIRIT OF ST. LOUIS		
	1951	1952	1953	1951	1952	1953
45-73.....	—	37.5	25.0	—	25.0	25.0
45-80.....	—	0.0	0.0	—	0.0	25.0
46-3.....	—	25.0	0.0	—	0.0	0.0
46-5.....	—	0.0	0.0	—	0.0	0.0
46-9.....	—	12.5	0.0	—	12.5	12.5
46-14.....	—	0.0	0.0	—	0.0	0.0
47-6.....	—	50.0	0.0	—	0.0	0.0
47-10.....	—	25.0	0.0	—	0.0	0.0
49-4.....	—	100.0	75.0	—	0.0	0.0
49-15.....	—	0.0	0.0	—	0.0	0.0
49-23.....	—	0.0	0.0	—	0.0	0.0
49-30.....	—	100.0	0.0	—	0.0	0.0
49-31.....	—	25.0	50.0	—	0.0	0.0
50-6.....	—	75.0	0.0	—	0.0	0.0
50-24.....	—	25.0	0.0	—	0.0	0.0
50-27.....	100.0	75.0	75.0	12.5	0.0	0.0
50-28.....	—	75.0	25.0	—	0.0	75.0
45-8.....	—	62.5	50.0	—	0.0	0.0
45-74.....	—	0.0	62.5	—	0.0	0.0
45-75.....	—	75.0	100.0	—	0.0	0.0
45-78.....	—	0.0	0.0	—	0.0	0.0
46-4.....	—	25.0	12.5	—	0.0	0.0
46-12.....	—	100.0	37.5	—	0.0	0.0
47-1.....	—	87.5	100.0	—	0.0	12.5
47-8.....	—	37.5	12.5	—	0.0	0.0
47-12.....	—	100.0	75.0	—	0.0	0.0
47-19.....	—	100.0	75.0	—	0.0	0.0
47-32.....	—	100.0	75.0	—	0.0	50.0
49-1.....	—	100.0	75.0	—	0.0	25.0
49-17.....	100.0	100.0	87.5	12.5	0.0	0.0
49-19.....	—	100.0	0.0	—	0.0	0.0
50-7.....	—	100.0	25.0	—	37.5	37.5
50-22.....	100.0	100.0	75.0	12.5	12.5	37.5
50-25.....	—	0.0	0.0	—	0.0	0.0
47-2.....	—	87.5	75.0	—	87.5	25.0
47-3.....	—	100.0	100.0	—	12.5	0.0
49-8.....	75.0	87.5	12.5	87.5	75.0	37.5
49-20.....	—	87.5	37.5	—	25.0	0.0
50-23.....	—	100.0	62.5	—	12.5	0.0
50-26.....	—	100.0	75.0	—	0.0	12.5

Table 18.—Results of greenhouse tests on gladiolus variety Picardy inoculated with *Fusarium*, 1948.

ISOLATE	SYMPTOMS AND NUMBERS OF CORMS AFFECTED			
	Vascular Discoloration	Basal Dry Rot	Completely Destroyed	No Symptoms
45-73.....	6	0	0	4
46-3.....	0	4	1	5
46-9.....	0	0	0	10
46-14.....	0	0	0	10
47-6.....	0	3	1	6
45-8.....	0	7	1	2
45-74.....	0	0	0	10
45-75.....	0	0	10	0
46-4.....	0	1	1	8
46-12.....	0	9	0	1
47-1.....	0	0	10	0
47-12.....	0	0	10	0
47-32.....	0	8	2	0
Check.....	0	0	0	10

differences in results, especially in the case of the variety Rosa Van Lima, which developed rot in 1951 but not in 1952. The over-all results seem to indicate that, as has been reported for many other pathogenic organisms, the isolates lose their virulence when maintained in laboratory culture over a period of time. More precise tests over a longer period of years would be necessary to establish this point with certainty.

Greenhouse Tests

In the greenhouse, corms were planted in soil which, after being steamed, had been infested with cultures of *Fusarium* that had been grown on autoclaved whole-grain oats in 250-milliliter Erlenmeyer flasks for 2 to 3 weeks. The flasks were shaken daily during the growth period of the *Fusarium*.

In order that the chances of using infected gladiolus corms in these experiments might be minimized, the husks were removed and the corms were soaked 15 minutes in a solution of 9 grams *New Improved Ceresan* (5 per cent ethyl mercury phosphate) in 1 gallon of water. The corms were then washed in several changes of tap water to remove the *Ceresan*. Corms used in some of the tests were treated in this same way.

Furrows 2½ to 3 inches deep were marked out across a greenhouse bench. Depressions 4 inches apart were made in

the bottoms of the furrows. One *Fusarium*-carrying oat grain was placed in each depression and covered lightly with soil. A gladiolus corm was then set over it. After the corms had been placed, the furrows were filled with soil.

No attempt was made to maintain a constant soil moisture content. Water was applied to the soil as often as necessary to maintain good growing conditions.

Tests in 1948.—Preliminary tests with 13 isolates of *Fusarium* were made in 1948. Ten Picardy corms, each approximately three-fourths inch in diameter, were used for each isolate. Planting was done March 1. All corms produced new plants. By April 19, yellow leaves indicated disease in some of the plants. New corms were examined August 5. Symptoms produced by the different isolates and the numbers of corms affected are shown in table 18.

Tests in 1952.—Twenty isolates of *Fusarium* were tested in 1952 for pathogenicity on the varieties Dr. F. E. Bennett, Margaret Fulton, Picardy, and Variation. Ten commercial size No. 5 (one-half to three-fourths inch in diameter) corms of each variety were used with each isolate. Three groups of checks, each consisting of 10 corms of each variety, were used in this experiment.

Planting was done on February 21 and 22. Differences in reaction of the four varieties to some of the isolates were quite pronounced by April 14, figs. 20 and 21.

The new corms were dug July 24; they were cleaned and examined August 13. They were then placed in a refrigerator

at 5 degrees C. and left until November or December, when isolations from the affected corms were attempted.



Fig. 20.—Differences in reaction of, left to right in each picture, gladiolus varieties Dr. F. E. Bennett, Margaret Fulton, Picardy, and Variation to basal dry rot isolate 49-20 and vascular isolates 49-4 and 50-6 of *Fusarium*. Photographs were taken 52 days after planting date.

Results obtained in these tests are shown in tables 19, 20, 21, and 22. In the cases of the Dr. F. E. Bennett and Picardy varieties, the results cannot be considered

conclusive, because disease symptoms developed in some corms of the checks. *Fusarium* was isolated from two corms of the Dr. F. E. Bennett checks. This finding

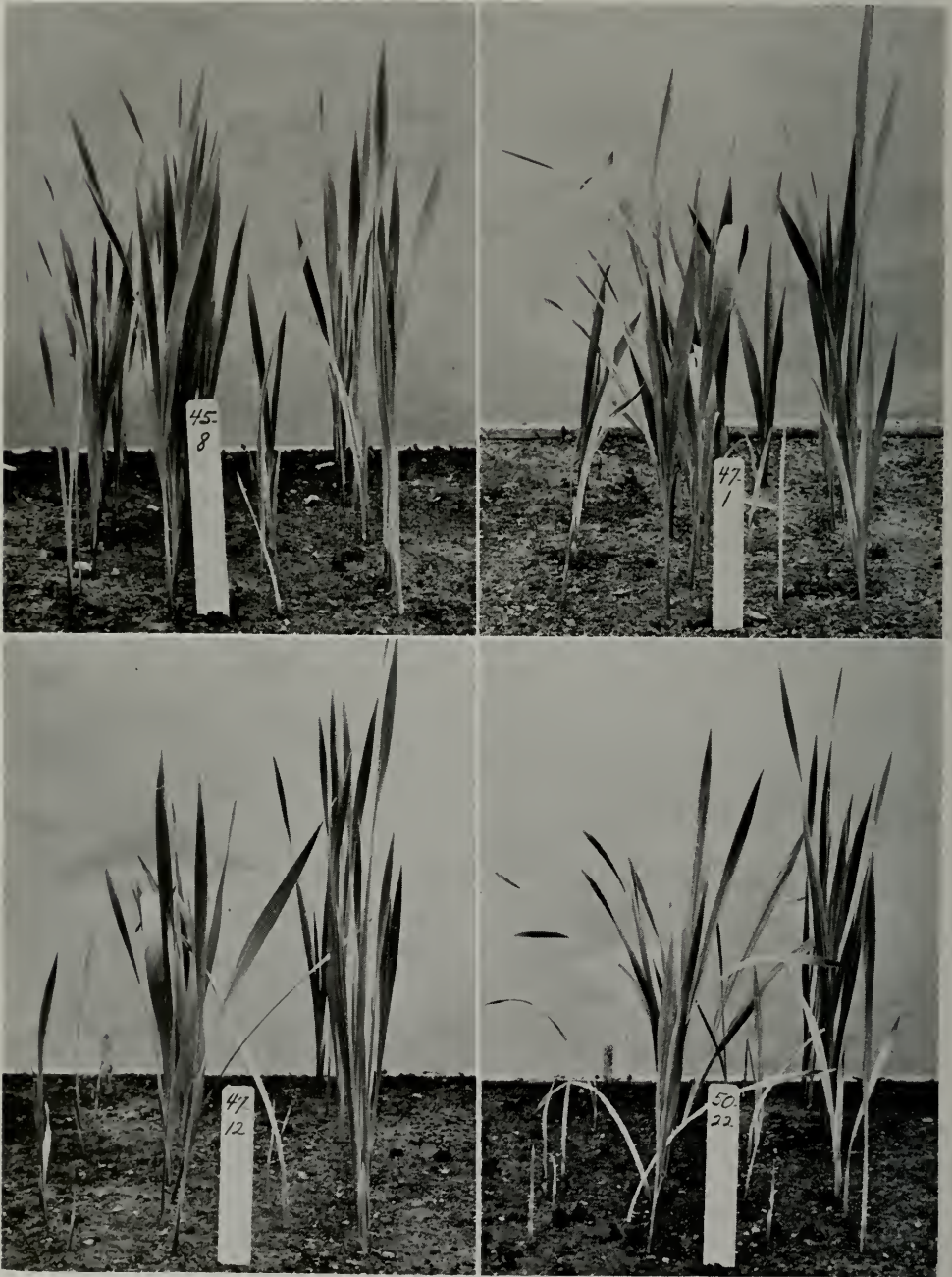


Fig. 21.—Differences in reaction of, left to right in each picture, gladiolus varieties Dr. F. E. Bennett, Margaret Fulton, Picardy, and Variation to four brown rot isolates of *Fusarium*. Photographs were taken 52 days after planting date.

indicated that the planting stock carried some *Fusarium*; so all of the symptoms that developed in the new corms probably did not result from the inoculations.

In the variety Picardy, five of the check plants died before reaching maturity and seven new corms had vascular discoloration. *Fusarium* was not isolated from any of these new corms. *Curvularia* sp. was isolated from four of them and *Penicillium* sp. was isolated from the other three. These two organisms appeared also in some isolations from corms in the inoculated series.

In the variety Variation, vascular discoloration developed in one corm; four corms of the 29 in the checks had slight basal rot lesions. Isolations from four of these affected corms yielded *Penicillium*; no organism was obtained from the fifth corm.

All plants from noninoculated Margaret Fulton corms remained healthy and

produced healthy daughter corms. *Fusarium* was recovered from diseased corms in the inoculated series 36 times in 52 attempts.

Tests in 1953.—Varieties Dr. F. E. Bennett, Margaret Fulton, Spotlight, and Elizabeth the Queen were inoculated late in 1952 with 13 isolates of *Fusarium*. Size No. 4 (three-fourths to 1 inch in diameter) corms of the variety Elizabeth the Queen and large cormels of the other three varieties were used in this test. Treating and planting was done December 29, 1952. New corms were harvested June 5, 1953; they were cleaned and examined 2 weeks later.

In this trial, because no disease was found in the new corms produced in the noninoculated checks, and because only one Dr. F. E. Bennett and one Spotlight plant failed to live and produce new corms, it was assumed that the planting stock was practically free of disease. Cause of death

Table 19.—Results of greenhouse tests on gladiolus variety Dr. F. E. Bennett inoculated with *Fusarium*, February, 1952; new corms were examined in August of the same year.

ISOLATE	PLANTS DEVELOPED	NUMBER OF NEW CORMS OBTAINED					FUSARIUM RECOVERED*
		Total	Healthy	Brown Rot	Vascular Discoloration	Basal Dry Rot	
Brown rot 45-8.....	10	9	2	0	7	0	3/8
Brown rot 45-74.....	10	8	8	0	0	0	0/0
Brown rot 47-1.....	9	0	0	0	0	0	0/0
Brown rot 47-12.....	8	0	0	0	0	0	0/0
Brown rot 47-32.....	10	5	0	0	5	0	5/5
Brown rot 49-8.....	10	10	4	0	6	0	1/5
Brown rot 50-7.....	10	8	6	0	2	0	2/2
Brown rot 50-22.....	7	0	0	0	0	0	0/0
Total.....	74	40	20	0	20	0	11/20
Vascular 45-73.....	10	6	0	0	6	0	3/4
Vascular 46-3.....	10	1	0	0	1	0	1/1
Vascular 46-9.....	10	10	0	0	10	0	0/0
Vascular 47-10.....	10	10	2	0	8	0	5/5
Vascular 49-4.....	9	0	0	0	0	0	0/0
Vascular 49-23.....	10	10	7	1	2	0	1/1
Vascular 50-6.....	8	0	0	0	0	0	0/0
Vascular 50-24.....	10	10	7	1	2	0	0/3
Vascular 50-28.....	10	5	0	0	5	0	0/0
Total.....	87	52	16	2	34	0	10/14
Basal dry rot 49-20.....	10	4	1	0	3	0	1/2
Basal dry rot 50-23.....	10	10	3	1	6	0	4/5
Basal dry rot 50-26.....	10	10	6	0	4	0	3/5
Total.....	30	24	10	1	13	0	8/12
Check No. 1.....	10	9	7	0	2	0	1/3
Check No. 2.....	10	10	7	0	3	0	1/3
Check No. 3.....	10	10	9	0	1	0	0/1
Total.....	30	29	23	0	6	0	2/7

*The numerator indicates the number of successful isolations, the denominator the number of corms from which isolations were attempted.

of the two nonsurviving check plants was not determined. As the results of this test are considered more conclusive than those of the previous test, they are given in detail by isolates and varieties.

Brown rot isolate 47-12

DR. F. E. BENNETT: Only 3 plants emerged; none produced a new corm.

MARGARET FULTON: 8 plants emerged; 1 survived and produced a new corm, which had thin basal rot.

SPOTLIGHT: 5 plants emerged; 4 survived and produced new corms, 3 of which had much of the bases rotted, but the rotted tissue was very thin. There was some vascular streaking, but it was not general. One corm showed no symptoms. *Fusarium* was recovered from all 3 affected corms.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. Four of the new corms were badly decomposed; 6 had typical brown rot le-

sions. Of these 6 corms, 1 had numerous brown vascular strands; another had a single brown vascular strand in addition to the brown rot lesions. *Fusarium* was recovered from all 6 corms.

Brown rot isolate 47-32

DR. F. E. BENNETT: 6 plants emerged; 5 survived and produced new corms, all of which had core rot and brown vascular streaks. *Fusarium* was recovered from all 5 corms.

MARGARET FULTON: 10 plants emerged; all survived and developed new corms. Two corms had thin basal rot lesions; 8 showed no symptoms. *Fusarium* was not recovered from the affected corms.

SPOTLIGHT: 5 plants emerged; 7 survived and produced new corms, all of which had diffused basal lesions. There was no vascular discoloration. Cultures were made from 3 corms, and *Fusarium* was recovered from each corm.

Table 20.—Results of greenhouse tests on gladiolus variety Margaret Fulton inoculated with *Fusarium*, February, 1952; new corms were examined in August of the same year.

ISOLATE	PLANTS DEVELOPED	NUMBER OF NEW CORMS OBTAINED					FUSARIUM RECOVERED*
		Total	Healthy	Brown Rot	Vascular Discoloration	Basal Dry Rot	
Brown rot 45-8.....	10	10	10	0	0	0	0/0
Brown rot 45-74.....	10	10	9	0	1	0	0/0
Brown rot 47-1.....	10	5	0	0	5	2	4/5
Brown rot 47-12.....	10	1	0	0	1	0	0/1
Brown rot 47-32.....	10	10	6	0	4	3	1/3
Brown rot 49-8.....	10	10	8	0	2	0	1/3
Brown rot 50-7.....	10	10	10	0	0	0	0/0
Brown rot 50-22.....	10	4	1	1	2	0	1/2
Total.....	80	60	44	1	15	5	7/14
Vascular 45-73.....	10	10	10	0	0	0	0/0
Vascular 46-3.....	10	9	3	0	6	0	3/4
Vascular 46-9.....	10	10	9	0	1	0	1/1
Vascular 47-10.....	10	10	9	0	1	0	1/1
Vascular 49-4.....	10	2	0	0	2	1	1/2
Vascular 49-23.....	10	10	10	0	0	0	0/0
Vascular 50-6.....	10	10	1	0	9	0	6/9
Vascular 50-24.....	10	10	7	0	3	0	0/4
Vascular 50-28.....	10	10	0	0	10	0	10/10
Total.....	90	81	49	0	32	1	22/31
Basal dry rot 49-20.....	10	10	0	0	10	0	5/5
Basal dry rot 50-23.....	10	10	8	0	2	0	2/2
Basal dry rot 50-26.....	10	10	10	0	0	0	0/0
Total.....	30	30	18	0	12	0	7/7
Check No. 1.....	10	10	10	0	0	0	0/0
Check No. 2.....	10	10	10	0	0	0	0/0
Check No. 3.....	10	10	10	0	0	0	0/0
Total.....	30	30	30	0	0	0	0/0

*The numerator indicates the number of successful isolations, the denominator the number of corms from which isolations were attempted.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms, 7 of which had very thin small lesions at the sides of the core bases. *Fusarium* was recovered from 2 of 4 corms cultured.

Brown rot isolate 50-7

DR. F. E. BENNETT: 8 plants emerged; all survived and produced new corms. All new corms had thin basal rot lesions. *Fusarium* was recovered from 2 of 5 corms cultured.

MARGARET FULTON: 10 plants emerged; all survived and produced new corms. No disease symptoms developed.

SPOTLIGHT: 10 plants emerged; all survived and produced new corms. Nine of the new corms appeared to have thin basal rot lesions developing when the corms were cleaned, but the symptoms were not distinct. The corms were placed in an incubator at 26 degrees C. and left

for 15 weeks. The lesions did not develop further, and attempts to recover the *Fusarium* from these corms failed.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms, 9 of which had no disease symptoms. The remaining corm had a diffused basal rot lesion spreading halfway up the side of the corm. *Fusarium* was recovered from this lesion.

Brown rot isolate 50-22

DR. F. E. BENNETT: 6 plants emerged; 5 survived and produced new corms. Four of the new corms had typical thick brown rot lesions. One corm also had brown vascular strands. *Fusarium* was recovered from all 4 affected corms. One corm had no disease symptoms.

MARGARET FULTON: 10 plants emerged; 8 survived and produced new corms. Seven of the new corms had brown rot lesions on the bases and sides. Four

Table 21.—Results of greenhouse tests on gladiolus variety Picardy inoculated with *Fusarium*, February, 1952; new corms were examined in August of the same year.

ISOLATE	PLANTS DEVELOPED	NUMBER OF NEW CORMS OBTAINED					FUSARIUM RECOVERED*
		Total	Healthy	Brown Rot	Vascular Discoloration	Basal Dry Rot	
Brown rot 45-8.....	9	1	0	0	0	1	0/0
Brown rot 45-74.....	10	3	2	1	0	0	1/1
Brown rot 47-1.....	6	1	0	0	1	1	0/1
Brown rot 47-12.....	4	0	0	0	0	0	0/0
Brown rot 47-32.....	10	2	0	0	0	2	0/0
Brown rot 49-8.....	10	9	6	0	3	0	2/6
Brown rot 50-7.....	10	2	2	0	0	0	0/1
Brown rot 50-22.....	7	1	0	1	0	0	1/1
Total.....	66	19	10	2	4	4	4/10
Vascular 45-73.....	10	9	3	0	6	0	6/6
Vascular 46-3.....	10	3	0	0	3	0	0/3
Vascular 46-9.....	10	10	10	0	0	0	0/0
Vascular 47-10.....	10	9	8	0	1	0	0/1
Vascular 49-4.....	7	0	0	0	0	0	0/0
Vascular 49-23.....	10	8	6	0	0	2	0/4
Vascular 50-6.....	10	9	5	2	2	0	0/4
Vascular 50-24.....	10	6	1	0	0	5	1/4
Vascular 50-28.....	9	8	1	0	7	0	7/7
Total.....	86	62	34	2	19	7	14/29
Basal dry rot 49-20.....	10	5	4	0	1	0	1/3
Basal dry rot 50-23.....	10	3	0	0	2	1	0/2
Basal dry rot 50-26.....	10	2	0	0	0	2	1/1
Total.....	30	10	4	0	3	3	2/6
Check No. 1.....	10	8	5	0	3	0	0/3
Check No. 2.....	10	8	4	0	4	0	0/3
Check No. 3.....	10	9	9	0	0	0	0/4
Total.....	30	25	18	0	7	0	0/10

*The numerator indicates the number of successful isolations, the denominator the number of corms from which isolations were attempted.

corms were cultured, and *Fusarium* was recovered from each of them. One corm remained healthy.

SPOTLIGHT: 10 plants emerged; 6 survived and produced new corms. Three of the new corms were completely mummified at cleaning time. One corm was three-fourths rotted; another contained a discolored vascular strand but had no external symptoms. *Fusarium* was recovered from the rotted corm but not from the discolored vascular strand. One corm remained healthy.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. Two of the new corms were mummified at cleaning time, the other 8 had no disease symptoms. No attempt was made to recover the *Fusarium*.

Vascular isolate 45-73

DR. F. E. BENNETT: 7 plants emerged; 5 survived and produced new corms. Two

of the new corms had brown vascular strands; 3 had no symptoms. *Fusarium* was recovered from both of the affected corms.

MARGARET FULTON: 10 plants emerged; all survived and produced new corms. One corm had brown vascular strands; 9 had no symptoms. *Fusarium* was recovered from the affected corm.

SPOTLIGHT: 10 plants emerged; all survived and produced new corms. Three of the new corms had brown vascular strands; 7 remained healthy. *Fusarium* was recovered from 2 of the affected corms.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. Eight of the new corms had extensive discolored vascular strands; 2 of them had thick brown rot lesions also. No connections between the brown rot lesions and the vascular streaks were found in any of the corms. *Fusarium* was recovered from all 8 of the affected corms. Two of

Table 22.—Results of greenhouse tests on gladiolus variety Variation inoculated with *Fusarium*, February, 1952; new corms were examined in August of the same year.

ISOLATE	PLANTS DEVELOPED	NUMBER OF NEW CORMS OBTAINED					FUSARIUM RECOVERED*
		Total	Healthy	Brown Rot	Vascular Discoloration	Basal Dry Rot	
Brown rot 45-8.....	10	7	2	0	5	0	0/4
Brown rot 45-74.....	10	8	6	0	2	0	0/0
Brown rot 47-1.....	10	3	2	0	1	1	1/1
Brown rot 47-12.....	10	6	0	0	6	5	2/4
Brown rot 47-32.....	10	6	0	0	6	6	3/5
Brown rot 49-8.....	10	10	3	0	7	0	4/7
Brown rot 50-7.....	9	7	0	0	0	0	0/0
Brown rot 50-22.....	10	3	1	1	1	0	0/0
Total.....	79	50	14	1	28	12	10/21
Vascular 45-73.....	10	9	1	0	8	0	5/6
Vascular 46-3.....	10	10	7	0	3	0	0/3
Vascular 46-9.....	10	10	6	0	4	0	3/3
Vascular 47-10.....	10	10	10	0	0	0	0/0
Vascular 49-4.....	10	8	0	0	1	7	0/0
Vascular 49-23.....	10	10	10	0	0	0	0/0
Vascular 50-6.....	10	10	7	0	3	0	0/2
Vascular 50-24.....	10	10	8	0	1	1	0/1
Vascular 50-28.....	10	9	0	0	9	0	1/3
Total.....	90	86	49	0	29	8	9/18
Basal dry rot 49-20.....	10	10	10	0	0	0	0/0
Basal dry rot 50-23.....	10	9	7	0	2	0	0/2
Basal dry rot 50-26.....	9	7	3	0	0	4	0/2
Total.....	29	26	20	0	2	4	0/4
Check No. 1.....	10	9	9	0	0	0	0/0
Check No. 2.....	10	10	6	0	0	4	0/4
Check No. 3.....	10	10	9	0	1	0	0/1
Total.....	30	29	24	0	1	4	0/5

*The numerator indicates the number of successful isolations, the denominator the number of corms from which isolations were attempted.

the 10 corms had no visible symptoms of disease.

Vascular isolate 46-3

DR. F. E. BENNETT: 7 plants emerged; all survived and produced new corms. One of the new corms was mummified; 2 had basal core rot but no extensive vascular discoloration. Four of the corms remained healthy. *Fusarium* was recovered from both corms with core rot. Isolations were not attempted from the mummified corms.

MARGARET FULTON: 9 plants emerged; all survived and produced new corms. One had a thin basal rot lesion; 8 remained healthy. *Fusarium* was not recovered from the affected corm.

SPOTLIGHT: 10 plants emerged; all survived and produced new corms. None had disease symptoms.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. All of the new corms remained healthy.

Vascular isolate 47-10

DR. F. E. BENNETT: 7 plants emerged; all survived and produced new corms. Three corms had thin basal core rot but no vascular discoloration. Four corms had no disease symptoms. *Fusarium* was recovered from 2 of the 3 affected corms.

MARGARET FULTON: 9 plants emerged; 8 survived and produced new corms. One had extensive discolored vascular strands; 7 remained healthy. *Fusarium* was recovered from the affected corm.

SPOTLIGHT: 10 plants emerged; all survived and produced new corms. All of the new corms remained healthy.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. All of the new corms remained healthy.

Vascular isolate 49-4

DR. F. E. BENNETT: 8 plants emerged; 7 survived and produced new corms. Two corms were mummified; 1 had a deep core rot extending to a thin lesion at the side of the core base. Three corms had deep core rot but no extensive vascular discoloration. One corm remained healthy. *Fusarium* was recovered from 3 of the affected corms.

MARGARET FULTON: 9 plants emerged; all survived and produced new corms. Eight corms had thin basal rot. There was no vascular discoloration. Three corms were cultured, and *Fusarium* was recovered from all of them. One corm had no disease symptoms.

SPOTLIGHT: 10 plants emerged; 9 survived and produced new corms. At cleaning time 2 of the corms were mummified; 6 had thin basal rot. *Fusarium* was recovered from 5 of the 6 corms affected with basal rot.

ELIZABETH THE QUEEN: 10 plants emerged; 9 survived and produced new corms. Four of the new corms had thin basal rot. In 1 corm, the brown discoloration extended from the base into one vascular strand. *Fusarium* was recovered from 1 of the 4 affected corms.

Vascular isolate 50-24

Eight DR. F. E. BENNETT, 9 MARGARET FULTON, 10 SPOTLIGHT, and 10 ELIZABETH THE QUEEN plants emerged; all survived and produced new corms. No disease symptoms developed.

Basal rot isolate 47-2

DR. F. E. BENNETT: 10 plants emerged; all survived and produced new corms. One of the new corms had one brown vascular strand; another had very short discolored vascular streaks around the core. *Fusarium* was not recovered from the affected corms. Eight corms had no disease symptoms.

MARGARET FULTON: 10 plants emerged; 9 survived and produced new corms. All new corms remained healthy.

SPOTLIGHT: 8 plants emerged; all survived and produced new corms. Six corms had thin basal rot lesions, most of which chipped out when the corms were cleaned. One corm also had pronounced vascular discoloration. *Fusarium* was recovered from 3 of the affected corms. Two corms remained healthy.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. All new corms remained healthy.

Basal rot isolate 47-3

DR. F. E. BENNETT: 7 plants emerged; all survived and produced new corms. All

7 corms had brown core bases; 5 of these corms had brown vascular strands also. *Fusarium* was recovered from all 7 corms.

MARGARET FULTON: 10 plants emerged; all survived and produced new corms. Each of the corms had a thin, light brown discoloration around the core base. Only 1 corm had a discolored vascular strand. Three of the corms were cultured; *Fusarium* was recovered from all of them.

SPOTLIGHT: 10 plants emerged; all survived and produced new corms. One corm had a hard core base and pronounced vascular discoloration. Two other corms had lighter vascular discoloration. *Fusarium* was not recovered from these corms. Seven corms showed no symptoms of disease.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. Four corms had typical thin basal rot lesions; 2 had discolored vascular strands but no basal rot lesions. *Fusarium* was recovered from 5 of the 6 affected corms. Four corms had no symptoms of disease.

Basal rot isolate 50-23

DR. F. E. BENNETT: 6 plants emerged; all survived and produced new corms. One corm had a thin basal rot lesion; 1 had a brown vascular strand in the top half of the corm. *Fusarium* was recovered from both affected corms. Four corms remained healthy.

MARGARET FULTON: 10 plants emerged; 9 survived and produced new corms. Eight corms had thin brown discolorations over the core bases. In 1 corm the discoloration extended slightly beyond the core. One corm had no symptoms. Attempts to recover *Fusarium* failed with all 8 affected corms.

SPOTLIGHT: 10 plants emerged; all survived and produced new corms. Nine corms had typical thin diffused basal rot lesions. There was no vascular discoloration. *Fusarium* was recovered from 8 of the 9 affected corms. One corm remained healthy.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. No disease symptoms developed in any of the plants or new corms.

Basal rot isolate 50-26

DR. F. E. BENNETT: 8 plants emerged; all survived and produced new corms. Seven corms had brown core bases. One corm also had a thick brown rot lesion on its side; 1 corm had a thin brown rot lesion on its side, and 1 corm had a brown vascular strand. One corm had no disease symptoms. *Fusarium* was recovered from 5 of the 7 affected corms.

MARGARET FULTON: 9 plants emerged; all survived and produced new corms. Six corms had brown core bases; 3 had no symptoms. *Fusarium* was recovered from only 1 of the affected corms.

SPOTLIGHT: 9 plants emerged; 8 survived and produced new corms. Two of the corms were mummies; 6 had typical thin basal rot lesions. *Fusarium* was recovered from all 6 of these corms.

ELIZABETH THE QUEEN: 10 plants emerged; all survived and produced new corms. Two corms had very small, thin basal rot lesions. *Fusarium* was recovered from both corms. Eight corms had no disease symptoms.

Noninoculated checks: Three sets of checks consisting of 10 corms of each variety in each set were planted.

DR. F. E. BENNETT: 24 plants emerged; 23 survived and produced new corms. No disease symptoms developed.

MARGARET FULTON: 29 plants emerged; all survived and produced new corms. No disease symptoms developed.

SPOTLIGHT: 29 plants emerged; 28 survived and produced new corms. No disease symptoms developed.

ELIZABETH THE QUEEN: 30 plants emerged; all survived and produced new corms. No disease symptoms developed.

Fusarium reisolates: The cultures that were recovered from the new corms agreed closely in growth type on Wellman's agar with the growth types of the respective isolates used for inoculation.

Variation in Virulence of Isolates.—Relative virulences of the isolates used in the greenhouse tests were rated numerically by means of disease indexes computed as follows: Values were assigned to the disease symptoms in the new corms and these values were applied in a modifi-

cation of the formula used in determination of rot severity indexes in the laboratory tests. Complete decomposition of the corms was considered the most severe result of the disease and was assigned a value

of 3. Brown rot and vascular symptoms were considered equal in severity and were assigned a value of 2. Thin basal dry rot was considered less severe and was given a value of 1. Absence of symptoms rated

Table 23.—Severity indexes (obtained by use of formula on page 496) for the disease produced by 20 isolates of *Fusarium* on four gladiolus varieties in greenhouse tests, 1952.*

ISOLATE	VARIETY				AVERAGE DISEASE INDEX
	Dr. F. E. Bennett	Margaret Fulton	Picardy	Variation	
45-8.....	56.1	0.0	92.6	63.3	53.0
45-74.....	20.0	6.7	76.7	33.3	34.2
47-1.....	100.0	90.0	94.4	80.0	91.1
47-32.....	80.3	36.7	86.7	80.0	70.9
49-8.....	40.0	13.3	30.0	46.7	32.5
50-7.....	33.3	0.0	80.0	30.0	35.8
50-22.....	100.0	80.0	95.2	76.7	88.0
45-73.....	80.0	0.0	50.0	63.3	48.3
46-3.....	96.7	50.0	80.0	20.0	61.7
46-9.....	66.7	6.7	0.0	26.7	25.0
47-10.....	53.3	6.7	16.7	0.0	19.2
49-4.....	100.0	96.7	100.0	50.0	86.7
49-23.....	20.0	0.0	26.7	0.0	11.7
50-6.....	100.0	60.0	36.7	20.0	54.2
50-24.....	20.0	20.0	56.7	10.0	26.7
50-28.....	33.3	66.7	63.0	70.0	58.2
47-12.....	100.0	96.7	100.0	70.0	91.7
49-20.....	80.0	66.7	56.7	0.0	50.8
50-23.....	46.7	13.3	86.7	23.3	42.5
50-26.....	26.7	0.0	86.7	43.3	39.2
Check No. 1.....	23.3	0.0	40.0	10.0	18.3
Check No. 2.....	20.0	0.0	46.7	13.3	20.0
Check No. 3.....	6.7	0.0	10.0	6.7	5.8

*Symptoms in all varieties except Margaret Fulton were due in part to organisms other than the isolates used for inoculation, as explained in the text.

Table 24.—Severity indexes (obtained by use of formula on page 496) for the disease produced by 13 isolates of *Fusarium* on four gladiolus varieties in greenhouse tests, 1953.

ISOLATE	VARIETY				AVERAGE DISEASE INDEX
	Dr. F. E. Bennett	Margaret Fulton	Spotlight	Elizabeth the Queen	
47-12.....	100.0	91.7	60.0	66.7	79.6
47-32.....	72.2	6.7	76.7	23.3	44.7
50-7.....	33.3	0.0	0.0	6.7	10.0
50-22.....	61.1	66.7	73.3	13.3	53.6
45-73.....	47.6	6.7	20.0	53.3	31.9
46-3.....	28.6	11.1	0.0	0.0	9.9
47-10.....	28.6	18.5	0.0	0.0	11.8
49-4.....	62.5	29.6	50.0	30.0	43.0
50-24.....	0.0	0.0	0.0	0.0	0.0
47-2.....	13.3	10.0	41.7	0.0	16.2
47-3.....	66.7	36.7	20.0	26.7	37.5
50-23.....	16.7	63.3	30.0	0.0	27.5
50-26.....	50.0	22.2	40.1	66.7	44.7
Check No. 1.....	0.0	0.0	0.0	0.0	0.0
Check No. 2.....	12.5	0.0	0.0	0.0	3.1
Check No. 3.....	0.0	0.0	10.0	0.0	2.5

a value of 0. The formula used was as follows:

$$\text{Disease index} = \frac{N_1 0 + N_2 1 + N_3 2 + N_4 3}{3t} \times 100$$

where N_1, N_2, N_3, N_4 = number of corms in each severity class

0, 1, 2, 3 = values assigned to severity classes

t = total number of corms counted

The disease indexes obtained from these calculations are shown in tables 23 and 24. The isolates varied in their effects on the test varieties, but the variations did not follow any definite pattern.

The results obtained in the inoculation tests show that an isolate does not always produce the same form of the disease. For example, in the 1953 tests, all three forms of the disease appeared in plants inoculated with isolates 47-32 and 50-26. In most cases, more than one disease form was obtained from inoculations with a single isolate. A summary of the disease forms produced in plants inoculated with the various isolates is given in table 25.

Although the same methods were not employed, these results are in general agreement with those obtained by McClellan (1948), who used mass culture isolates of *Fusarium* for inoculations on the Picardy and Dr. F. E. Bennett varieties. Some isolates caused a vascular rot, some a surface rot, and others caused a combination of symptoms. McClellan mentioned that the pathogenicity of isolates varied from year to year but he did not describe the nature of the variation.

DISCUSSION AND CONCLUSIONS

The occurrence of different forms of a single fungus in laboratory cultures has been reported by many workers in many parts of the world. The terms "variation," "saltation," "dissociation," "mutation," and "sectoring" have been used to designate this phenomenon.

Most of the studies of variations in the genus *Fusarium* have been made on the relation of pathogenicity to forms of growth. Most of the workers have re-

ported that cultures of *Fusarium* vary between a type with abundant aerial growth and a type with all the mycelium appressed in the nutrient substrate. The type of culture having aerial mycelium generally has been found to be most pathogenic, the type having only appressed mycelium least pathogenic, but observers on this point have not agreed unanimously (Armstrong, MacLachlan, & Weindling 1940, Burkholder 1925).

Numerous attempts have been made to account for these variations, but none has been entirely satisfactory. Leonian (1929, 1932) considered different cultural forms merely as phases in an orbit of variation of a species. Hansen (1938) explained variability in many of the Fungi Imperfecti as a "dual phenomenon" resulting from a segregation of genetically different nuclei in a multinucleate mycelium. Hansen & Smith (1932) earlier concluded that variable forms of the Fungi Imperfecti may owe their instability to nuclear heterogeneity, and that this condition can be brought about by nuclei of one strain entering the cells of another strain through anastomoses, and that reassortment of diverse nuclei can be accomplished by such mechanisms as anastomosis and unequal cell division.

Anastomoses, or fusions, between the hyphae and between the germ tubes of various fungi have been described by other workers. Zeller (1926) found conjugating macroconidia of *Nectria sanguinea* (Sibth.) Fr. in sporodochia on apple bark. He observed some comparatively long conjugating tubes connecting cells of one spore with those of another. In each case the nucleus had migrated from one cell to the other, producing a binucleate condition. Also, Zeller saw conjugation between two cells of the same spore. He did not discuss the significance of these conjugations.

Dickinson (1932) studied hyphal fusions in *Fusarium fructigenum* (Fries) and *F. vasinfectum* (Atk.). In no case did he observe fusion between segments (cells) of a conidium and only once did he find fusion between two segments of adjacent conidia. He concluded that in these species saltation was due to mutation rather than to heterocaryosis or cytoplasmic inheritance.

That germinating spores of the gladiolus *Fusarium* anastomose was observed in the studies reported here, fig. 22, but cytological studies were not made and no specific attempt was made to relate this phenomenon to subsequent variation of the isolates. If, however, the interpretation

of Hansen & Smith (1932) is correct, this phenomenon could account for the variants both in form of growth and in pathogenicity of the gladiolus *Fusarium*.

Miller (1945a, 1945b, 1946a, 1946b), in studies of the muskmelon-wilt *Fusarium* and other species, reported that when

Table 25.—Disease forms produced by 27 isolates of *Fusarium* in experiments performed in 1, 2, or 3 years.

ISOLATE	DISEASE FORM FROM WHICH ORIGINAL ISOLATE WAS OBTAINED	YEAR		
		1948	1952	1953
45-73.....	Vascular	Vascular	Vascular	Vascular, brown rot
46-3.....	Vascular	Basal dry rot	Vascular	Vascular, basal dry rot
46-9.....	Vascular	None	Vascular	—
46-14.....	Vascular	None	—	—
47-6.....	Vascular	Basal dry rot	—	—
47-10.....	Vascular	—	Vascular	Vascular
49-4.....	Vascular	—	Complete rot, vascular, basal dry rot	Basal dry rot, vascular
49-23.....	Vascular	—	Vascular	—
50-6.....	Vascular	—	Complete rot, vascular, brown rot	—
50-24.....	Vascular	—	Basal dry rot, vascular, brown rot	None
45-8.....	Brown rot	Basal dry rot	Vascular	—
45-74.....	Brown rot	None	Vascular, brown rot	—
45-75.....	Brown rot	Complete rot	—	—
46-4.....	Brown rot	Basal dry rot	—	—
46-12.....	Basal dry rot	Basal dry rot	—	—
47-1.....	Brown rot	Complete rot	Complete rot, vascular, basal dry rot	—
47-12.....	Brown rot	Complete rot	Complete rot, vascular, basal dry rot	Complete rot, brown rot, vascular
47-32.....	Brown rot	Basal dry rot	Vascular, basal dry rot	Brown rot, vascular, basal dry rot
50-7.....	Brown rot	—	Complete rot	Basal dry rot, brown rot
50-22.....	Brown rot	—	Complete rot, brown rot, vascular	Brown rot, vascular
47-2.....	Basal dry rot	—	—	Basal dry rot, vascular
47-3.....	Basal dry rot	—	—	Basal dry rot, vascular
49-8.....	Brown rot	—	Vascular	—
49-20.....	Basal dry rot	—	Vascular	—
50-23.....	Basal dry rot	—	Vascular, basal dry rot, brown rot	Basal dry rot, vascular
50-26.....	Basal dry rot	—	Basal dry rot, vascular	Basal dry rot, brown rot, vascular
50-28.....	Vascular	—	Vascular	—

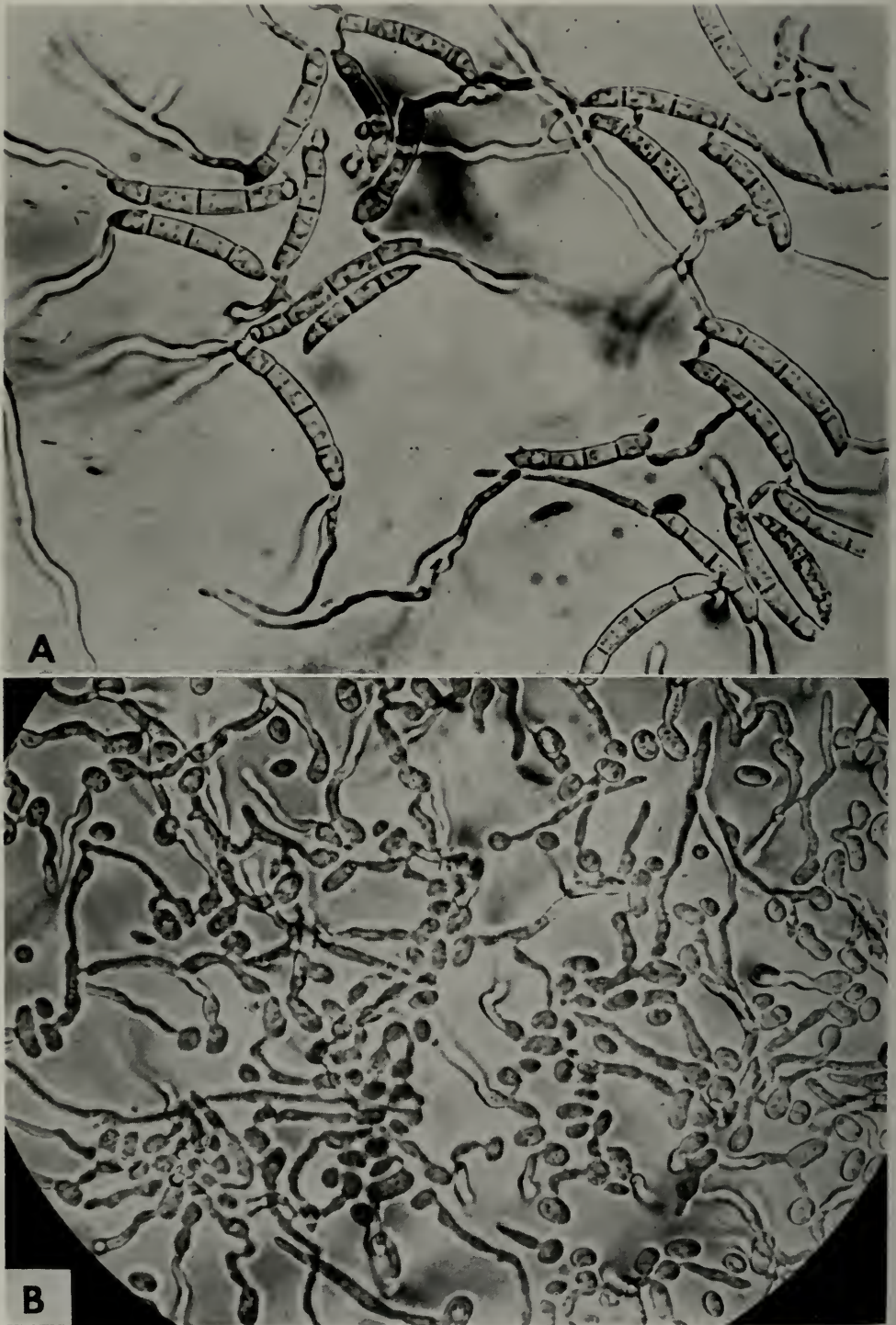


Fig. 22.—Anastomoses among germinating spores of the gladiolus *Fusarium*: A, macrospores; B, microspores.

monosporous isolations were made from an isolate derived from a diseased plant, all cultures were alike and of a form that produced abundant aerial mycelium on which conidia, mostly nonseptate, were borne rather sparsely. He called this form the wild type. Cultural variants that were derived from the wild type were considered to be laboratory mutants. Miller contended that the taxonomy of *Fusarium* should be based only on wild types and not on laboratory mutants.

The classification of Wollenweber & Reinking (1935) is based on the view that the macroconidia are the spore types on which species descriptions should be based and that those *Fusaria* which do not produce such spores when isolated from nature may be induced to do so by frequent cultural transfers. Such transferring gives rise to a cultural state characterized by abundant production of macroconidia, a condition that has been called "normal culture," "high culture," "Normkultur," and "Hochkultur." Miller contended that this method results in displacement of the wild type by mutants. According to Miller, most of the species descriptions appearing in the literature have been based on laboratory mutants rather than on natural types. Hence, he considered the classification of Wollenweber & Reinking unsatisfactory. The Snyder & Hansen (1940, 1941, 1945) revision disregards morphological criteria and places primary emphasis on the host relationship. Miller contended that this system also is unsatisfactory, stating that if proper cultural methods are employed the morphology of these organisms will be found sufficiently constant to warrant an attempt at morphological classification. He based this statement on two facts: (1) his original isolates were all of the raised type and (2) he was able to maintain this type by a soil culture technique in which the fungus was kept in a dried, inactive condition for long periods of time. From these two facts he concluded that variants of the types found in laboratory cultures do not occur in nature, or at least that variants are very rare. These conclusions do not agree with the report of Orton (1935), who found that several strains of *Fusarium niveum* Smith may

change as readily in soil as upon laboratory media.

While an extensive study of the occurrence of changes in culture form of the gladiolus *Fusarium* was not made by the writer, such changes were observed, table 10. Since the raised, appressed, and intermediate forms were obtained in original isolations of the gladiolus *Fusarium*, it must be assumed that all of these forms occur in nature and that the wild types of this fungus are not all of one form.

The main object of this investigation was to determine if strains of *Fusarium* obtained from gladiolus corms having different disease symptoms could be fitted into well-defined groups on the basis of their pathogenicity and physiological characters. The isolates used in these studies do not fall into well-defined groups. Also the pathogenicity tests show that a single isolate is capable of producing more than one form of disease. Evidence obtained in the other tests in this investigation shows that the strains of the gladiolus *Fusarium* are extremely variable and some apparently are quite unstable. No definite pattern for association of the many variables could be determined; the variations seem to occur independently.

It was not intended that this paper should enter the controversy on the relative merits of the two available systems of classifying *Fusaria*. But, as both systems have been used in previous studies of the *Fusarium* disease of gladiolus, and at least two specific names under one system have been used for strains of the fungus associated with different forms of the disease, it becomes necessary to take a definite stand regarding the nomenclature and taxonomy of the gladiolus *Fusarium*. Since this investigation failed to establish the existence of well-defined strains of a *Fusarium* associated with the different forms of the disease, it seems most logical to regard all variants as members of a single species. Because the system of Snyder & Hansen is better suited for classification of this type of organism, it is proposed that all forms of the gladiolus *Fusarium* be included under the name *Fusarium oxysporum* f. *gladioli* (Massey) Snyder & Hansen.

SUMMARY

Three forms of the *Fusarium* disease of gladiolus, known as the vascular, brown rot, and basal dry rot forms, have been described by other workers. The agent or agents which cause these disease forms have been assigned various specific names, with the result that the exact relation of the different symptom types and their causal agent or agents has been in a state of confusion.

The purpose of this investigation was to rectify the confusion by determining if strains of *Fusarium* producing different symptoms could be fitted into well-defined groups on the basis of their pathogenicity and physiological characters.

From several hundred isolates of *Fusarium* that had been cultured from diseased gladiolus corms, 40 isolates were selected for comparison in pathogenicity tests and physiological studies. Comparisons of isolates from the three disease forms were made by means of their reactions to temperature, reactions to aniline

dyes, reactions to copper salts, reactions to mercuric chloride, color reactions on steamed rice, growth types on differential media, pH changes produced in liquid media, spore measurements, and tendency to reproduce the same or different disease forms in inoculated plants and corms.

The isolates varied a great deal in their reactions in these tests, but no definite pattern for association of variables could be determined; the variations seemed to occur independently. The isolates did not fall into well-defined groups; the isolates from the three disease forms could not be distinguished by any of the tests used. The pathogenicity tests showed that a single isolate is capable of producing more than one form of the disease.

The evidence obtained in these studies shows that strains of the gladiolus *Fusarium* are extremely variable and that some of them apparently are quite unstable. It is proposed that all forms of the gladiolus *Fusarium* be included under the name *Fusarium oxysporum* f. *gladioli* (Massey) Snyder & Hansen.

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I N D E X

The following index covers Articles 2, 3, 4, 5, and 6 of Volume 26 of the ILLINOIS NATURAL HISTORY SURVEY BULLETIN. An index of Article 1, *The Mayflies, or Ephemeroptera, of Illinois*, by B. D. Burks, begins on page 211 of this volume. Indexing of Articles 2, 3, 4, 5, and 6 has been limited for the most part to the names of birds, fish, insects, mammals, and plants mentioned in the articles. In most cases, the singular form of the word has been used in the index, even though the plural form has been used in the text, as *fungus* for both *fungus* and *fungi*. Place names have not been indexed.

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