FUSARIUM WILT OF MUSKMELON

AND WATERMELON IN

SOUTHWESTERN ONTARIO.

# SOME ASPECTS OF THE <u>FUSARIUM</u> WILT OF MUSKMELON AND WATERMELON IN SOUTHWESTERN ONTARIO.

Ву

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TITIE: Some Aspects of the Fusarium Wilt of Muskmelon and Watermelon

in Southwestern Ontario.

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SCOPE AND CONTENTS: Distribution of Fusarium wilt of muskmelon and watermelon in southwestern Ontario was studied. Particular attention was paid
to morphological and physiological variations of the isolates obtained.

Morphological variations were based on comparison in culture with a selected
standard. Physiological variations were detected by pathogenicity experiments, and a study of assimilation of various carbon and nitrogen compounds.

Some further aspects of the biology of the organisms were investigated.

An experiment was carried out, employing several muskmelons and watermelon
varieties, to compare their resistance under field conditions.

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#### INTRODUCTION

In recent years the <u>Fusarium</u> wilts of muskmelons and watermelons have become increasingly important in Ontario. The Aldershot area, near Hamilton, and the extreme southern part of Essex county are the two main areas in which melons are grown. Commercial growing of watermelons in Essex county and the Port Dover area has been seriously affected, while muskmelon growing has been abandoned on many farms from Essex county to Dixie (near Toronto). This seriously affects the growing of both melons in Ontario, since the light sandy soil and high temperatures which they prefer, are found in very few places in the province.

The first record of Fusarium wilt of muskmelons in Ontario was made in 1936. In that year, the disease was observed in the Niagara Peninsula, and the grower reported that it had been present 2 or 3 years previously. It was reported by Chupp (2) in 1930 and 1931 as occurring in New York state, and in 1933 by Leach (10) in Minnesota. Not until 1943, according to McKeen (13), was it found in the principal melon growing area of Essex county. The disease has since spread rapidly and is now present in all major muskmelon growing districts in Ontario.

Fusarium wilt of watermelon was first described by Smith (21) in 1899 as occurring in South Carolina. He named the pathogen <u>Fusarium</u> niveum. McKeen (13) reports that <u>Fusarium</u> wilt was not observed attacking this host in the Essex area until 1948, although it had been common in watermelon growing areas of the United States for nearly 60 years. Since 1948 the disease has become very important in the area extending from

Harrow to Port Dover. It is interesting, however, that in the course of this investigation conversations with several growers, particularly one near Port Dover, indicate that the wilt was present much earlier.

The field symptoms of muskmelon involve stunting, wilting and death of the vines, but when less severe the vines survive and the fruit is small, bitter, immature and unmarketable. The symptoms of watermelon wilt involve death of the vine and immature or unpalatable fruit when the vines survive.

The causal organisms have long since been established as specie of <u>Fusarium</u>, which are designated as Forms 1 and 2 of <u>Fusarium bulbigenum var. niveum</u> Leach and Curence. Leach and Curence showed that the fungi causing wilt in watermelons and muskmelons were closely related, though they differed strikingly in pathogenicity. They concluded that the fungus causing watermelon wilt would not attack muskmelons and that the fungus causing muskmelon wilt would not attack watermelons.

The initial purpose in this investigation was to study the occurrence and distribution of the Fusaria causing wilt of watermelons and muskmelons in Ontario. After this study had been started it was found that more than one strain of both the muskmelon and watermelon pathogens existed. In view of this, experiments were undertaken to compare the pathogenicity of these isolates, and to determine whether or not their morphological differences could be correlated with their pathogenicity. In view of Gordon's (5) finding that two mating types existed in the Fusarium diseases of certain cereals, investigations to determine whether opposite mating types existed among the strains isolated were carried out.

Since little is known of the physiological differences between

different strains of pathogenic Fusaria other than the effect on host plants, attempts were made to correlate strain differences with the assimilation of various carbon and nitrogen sources.

#### MATERIALS AND METHODS.

Cultural media employed were Czapek Solution agar, Nitrogen base media, and Carbon base media. Czapek Solution agar, was the medium used in the classification of the various isolates, and in all places where a solid medium was needed. It gave more pronounced cultural differences between strains than did Potato Dextrose agar. Many isolates when compared on Potato Dextrose agar appeared identical, but when compared on Czapek agar these isolates would be different morphologically.

When comparing the morphology in culture of various strains on plates of Czapek agar acidified with 10% Lactic acid, each plate was inoculated four times, twice with the standard strain and twice with the strain to be compared with the standard. Each point of inoculation was 90° from the adjacent points. The plates were then incubated at 27°C. for 1 to 2 weeks after which time comparisons were made: (1) between the standard strain and the other strain on the plate and (2) between the various plates. If all cultures of strain 1 looked identical it was assumed that conditions were sufficiently uniform throughout the plates.

In the experiments investigating the utilization of carbon and nitrogen compounds liquid media were used. <u>Fusarium</u> growing in a liquid medium forms a thick mat-like layer on the surface and does not grow through the medium. For carbon utilization tests, the Nitrogen base medium of Wickerham (24) was employed, as prepared by the Difco laboratories. It is used in the classification of yeasts and contains all the essentials for yeast growth except a source of carbon. In preliminary

experiments it was found suitable for investigating the utilization of carbon sources by <u>Fusarium</u>. Since the only carbon source present is the one added, if growth occurs, then the carbon source has been assimilated. The selected carbon source was added in 1% concentration, and checks containing no carbon source were included for comparison.

The liquid medium for nitrogen assimilation was based on Wickerham's (24) Carbon base medium, but only the trace elements, salts, and glucose were used. The complete medium was not used as it was found that the <u>Fusarium</u> could grow in the Carbon base medium, even though a source of nitrogen was not added. This indicated the organism could utilize either the vitamins or amino acids present in the Carbon base medium as nitrogen sources.

Soil was infested by incorporating 1% of cornmeal-sand medium on which the organism had grown for 10 days. The ingredients of this medium were as follows: 1000 grams of sand: 200 grams of cornmeal: 250 mls. of water dispensed in 100 ml. quantities in 250 ml. flasks. Flasks were inoculated directly from stock cultures of the organisms maintained in soil tubes as described by Miller (17).

The soil used in the pathogenicity trials was a compost containing 3 volumes of sifted black loam, 2 volumes of fine sand, 1 volume of commercial fertilizer. This was placed in 10 litre flats and commercial peat moss added in the volume of 1%. This was thoroughly mixed, and these flats were used for planting. The soil in these trials was not sterilized. It was felt that addition of <u>Fusarium</u> to sterilized soil would give the organism an advantage which it does not have under normal field conditions. One hundred and twenty seeds were planted per flat:

40 Bender's Surprise muskmelon, 40 Iroquois muskmelon, and 40 Peerless watermelon. These varieties were selected for comparison since it gave pathogenicity comparisons for susceptible (Bender's Surprise), and resistant (Iroquois) muskmelon varieties, as well as indicating whether or not isolates from muskmelon could attack watermelon, and vice versa. Bender's Surprise is a large-fruited, vigorous melon with coarse, heavy vines. It is known to be very susceptible to <u>Fusarium</u> wilt. The Iroquois melon is a resistant variety developed by Munger (18) and released in 1943. It was selected from the cross of a <u>Fusarium</u> resistant line of melons similar to Golden Gopher with the Bender's Surprise variety. The vines are strong and vigorous, with round to oval deeply rubbed fruits, and deep orange flesh. The Peerless watermelon is a well known variety, with medium size vines, oblong, medium-green, fine-veined fruits, with bright red flesh.

Cultural purifications were made by flooding Czapek agar plates with conidial suspensions in sterilized distilled water. Excess water was drained by inverting the dish. Single spores were then removed by the "biscuit cutter" method of Keitt (8). No spatula was found necessary.

#### EXPERIMENTAL

 The Occurrence and Distribution of Strains of the <u>Fusarium</u> Causing Wilt of Muskmelon and Watermelon.

During this study some 369 isolates of <u>Fusarium</u> were obtained from melon fields. The location of these fields extended from Dixie (near Toronto) to Barrow. Ontario.

These isolates were obtained by (1) plating tissue from plants thought to be diseased and (2) planting seeds in soil obtained from fields believed to be infested with the disease, and making isolations from the seedlings which wilted. A tube of sterilized soil was inoculated with each isolate as soon as possible. These served as stock cultures for comparison experiments. This technique, as described by Miller (16), was used to keep the isolate free from mutation.

Comparison experiments were performed in two sections: (1) in which all isolates from muskmelon were compared against a chosen standard and (2) in which all isolates from watermelon compared against a chosen standard. The muskmelon standard was kindly supplied by Dr. J. J. Miller of McMaster University and is listed in the "Directory and Catalogue of Collections of Microorganisms maintained in Canada", issued by the National Research Council in 1951 as F23, Department of Botany, McMaster University. The watermeon standard was also supplied by Dr. J. J. Miller and is listed as F14, in the Catalogue. The "standard" cultures will be designated as strain I muskmelon wiit <u>Fusarium</u> and strain I watermelon wiit <u>Fusarium</u> respectively, in the present study.

# (A) Morphological Comparison of 270 Isolates of <u>Fusarium</u> from Muskmelon.

All isolates obtained from muskmelons were compared in this experiment. Since collections were made at different times, the comparison was made in 2 sections. In the first comparison 56 isolates were studied. The results are summarized in Table I.

The great majority of the isolates appeared identical with strain 1. However, 8 other types were found comprising 21% of the total. These all had abundant aerial mycelium and were not oppressed or deeply pigmented types.

In the second comparison experiment the remaining 214 isolates were compared strain 1. The results are listed in Table II. It is evident that once again a variety of isolates was obtained. Of the total of 214 isolates tested 112 were identical with strain 1. The other 102 or 47.6% are grouped in 9 other classes. Some of the isolates could not be fitted into these groups and these are listed as "others" in the table. The strains ranged from those very similar to strain 1 to brown types, resembling certain common saprophytic Fusaria. Small variations were noted within some strains, the distinctions not being quite as sharp as the table would indicate.

It should be noted that strain I was distributed over the entire sampled area, although it was not found on every sampled plot. Certain other strains were only sparsely distributed. For example, strain 9 was found only as 3 isolates, 2 from Dixie, and one from St. Thomas. Strain 2, however, was found in 17 isolates from the Aldershot area, and 3 from Dixie. Study of the tables will show further variations in abundance and distribution.

TABLE 1: Comparison of 56 Isolates of Fusarium from Muskmelon.

DISTR	IBUTION				STRAI	N				
AREA	FARM	1	A	В	С	D	E	F	G	H
Aldershot	Gallagher	20		1	1		1		1	
	Filman	9	1					2	1	
Leamington	Mastronardi	4	V							1
Ruthven	Colasanti	3								
Harrow	Duransky	4								
	llutz /	2	1							
	Tingen J.D.	2	1			1				
	Totals	44	3	1	1	1	1	2	2	1

TABLE II: Comparison of 214 Isolates of Fusarium from Muskmelon.

DISTRI	BUTION		1	NUSK	MELO	V ST	RAIN	NUMBER			
AREA	FARM	1	2	3	4	5	6	7 8	9	10	Others
Aldershot	Crouchley	20	7				1			1	
	DeLucca	3		1							1
	Filman	7	1	1		1					
	Gallagher	9	7	1						2	1
	Ricci	7	2	3	1	1		1			1
Dixie	Death	17							1		1
	McCarthy	13	3	2	2	1	2		1		1
Millgrove	Goodbrand					1					
Boston	Russling	5		2		1	1				2
Port Dover	Thompson	3		2	1	1					4
Aylmer	Howe	2					1				
Norwich	Mitro			1	2						
St.Thomas	Ward	2		6	1						
Harrow	Waters	10						2	1		2

TABLE II: (Continued)

DISTRIBU	TION	4	MU	SKME	LON	STR	AIN	NUMB	ER			
AREA	FARM	1	2	3	4	5	6	7	8	9	10	Others
	Plant							•				
	Breeding					1		2				
	Plot *											
	Station						1	3				
	Plot **											
	Tingen 0					1		1				4
	Wright R.	7										3
Ruthven	Colasanti	6	do	2					1			
Leamington	Mastronardi	1		1					2			
	Totals	112	20	22	7	8	6	6	6	3	3	20

<sup>\*</sup> Dominion Experimental Station.

<sup>\*\*</sup> Laboratory of Plant Pathology.

Another point worthy of note, however, is that certain sampled farms yielded a wide number of strains (McCarthy, near Dixie), while others yielded only a few strains (Death, near Dixie).

(B) Morphological Comparison of 99 Isolates of <u>Fusarium</u> from Water-melon.

The isolates tested in these experiments were obtained from watermelon — growing areas by the two methods mentioned for obtaining isolates
from muskmelon and the comparisons were made in the same manner. Strain 1
which was previously mentioned, was used as the standard. The results are
listed in Table III.

The total number of strains detected was 18. These showed similar variation to the muskmelon strains. However, of the 99 isolates tested, only 18, or 18.2% were like strain 1. Strain 1 was widely distributed, being found on most of the farms sampled. Strain 2 was obtained in as many isolations as strain 1, but its distribution was sparse, being localized almost entirely to one farm from the Harrow district. It would seem that this organism differs from the muskmelon pathogen in that on the whole, no one strain predominates.

It is interesting to speculate on the natural variation that has been found.

Miller (14) in previous work with muskmelons found only one type of isolate from the field, the strain 1 of these experiments. One would hestitate to attribute the multiplicity of strains observed here to mutation in culture since care was taken to minimize mutation by prompt transfer to soil tubes. Thus it is necessary to explain the difference in results between those of Miller and the writer. Several explanations may be suggested:

TABLE III: Distribution of 99 Isolates from Muskmelon.

DISTRI	EUTION			WAT	ERME	LON	STRA	IN N	UMBE	R		
AREA	FARM	1	2	3	4	5	6	7	8	9	10	Others
Boston	Russling	2		3								
Port Dover	Thompson	7	2	3			1					3
Aylmer	Howe	,				1		3	1			6
Norwich	Mitro	1				1	1		2	2		1
St.Thomas	Ward	2	1	1					1	1	1	1
Morpeth	Wade	1		1	1	2						2
Clear Creek	Alton	1			1	3					1	2
Harrow	Tingen 0.	1			3							1
	Tingen J.D.	1		1	2		1			1	1	1
	Wright R.	2	3			2		1				
	Mucola		13					2				
	Totals	18	19	9	6	5	8	6	4	4	3	17

(1) the experience of the present studies showed that strain differences tend to be more evident on Czapek Solution agar than on Potato Dextrose agar, the medium used by Miller in his work. Czapek Solution agar was used by the writer. (2) It was found that strain differences became more evident in older cultures than in cultures about I week old, and in this work comparisons were made in the 2-4 week period after inoculation, (3) other wild types may have become more abundant since the earlier work was done.

## 2. Experiments on the Pathogenicity of Some of the Isolates Obtained from Matermelon and Muskmelon.

These experiments were carried out to (1) determine whether individual strains vary in pathogenicity, and (2) to see if pathogenicity can be correlated with morphology.

One hundred and one of the 369 isolates obtained were included in these experiments. The isolates tested were selected from all 20 of the morphological types previously mentioned.

It is evident from Tables IV to VII that the isolates varied in their ability to attack the seedlings. For example isolate 298 listed in M2 (Table IV) caused 81.2% disease in Bender's, 0% in Peerless, and 65.5% disease in Iroquois. Similarly isolate 84 listed in M8 (Table IV) caused 40% disease in Bender's, 29.6% disease in Peerless, and 65.3% disease in Iroquois. In those cases where no disease incidence is apparent, it is probably safe to assume the <u>Fusarium</u> isolates under test here were sapprophytic types, originally obtained from weakened host plants.

Tables IV and VII list the results of a duplicated experiment to determine whether results obtained at one period of the summer could be repeated at a later date. The two experiments were begun 95 days apart.

TABLE IV: Pathogenicity of 18 Isolates from Muskmelon and 18

Isolates from Watermelon to Three Host Varieties.

STRA	ISOLATE	% GE	RM INAT	ION	% DISE	ASED 6	DAYS	% DIS	EASED 1	8 DAYS
-IN		В	P	I	В	P	1	В	P	1
M2	298	80	82.5	72.4	53.1	0	37.9	81.2	0	65.5
	280	70	77.5	77.5	39.3	0	32.2	100	0	96.7
м3	195	47.5	65	45	36.8	0	33.3	100	0	100
	176	70	70	62.5	0	0	0	0	0	0
M4	194	80	87.5	80	3.1	0	0	50.0	5,7	43.7
	215	50	52.5	3 <b>7.</b> 5	0	0	0	90.0	0	66.6
M5	147	88.1	82.6	82.6	0	0	0	0	0	0
	375	97.5	81.3	80.5	0	0	0	0	0	0
M6	256	97.5	90	85	0	0	0	0	0	0
	355	50	50	45	0	0	0	0	0	0
M7	354	92.5	87.5	82.5	0 *	0	0	0	0	0
	374	90.3	84.1	80.5	0	0	0	0	0	0
M8	84	75	67.5	65	0	0	0	40	29.6	65.3
	283	76	70	<b>7</b> 5	0	0	0	38.4	30	70.1
М9	98	65	82.5	65	2.5	0	2.5	42.3	6.1	25
	127	75	77.5	80	16.7	0.	.95	42.3	6	45
M10	135	47.5	60	22.5	26.3	0	11.1	73.6	0	88.8
	264	95	75	77.5	7.9	0	35.5	76.7	0	77.4
W2	336	72.5	57.5	62.5	0	47.9	0	3.4	91.3	8
	385	77.5	85	55	0	38.2	0	0	85.2	0
w3	173	100	62.5	87.5	0	0	2.9	10	8	8.6
	382	87.5	80	65	0	0	1	9	6	8.1
W4	352	97.5	82.5	75	0	0	0	46.1	21.2	50
	365	75	77.5	72.5	0	0	0	46.6	25.8	51.7

TABLE IV: (Continued)

STRA	ISOLATE	% GE	RM INAT	ION	% DI	SEASED 6	DAYS	% dis	EASED 1	6 DAYS
-IN		B	P	1	В	P	1	B	P	I
<b>35</b>	159	77.5	70	45	0	0	0	0	0	0
	216	87.5	80	87.5	0	0	0	O	0	0
Wó	168	65	02.5	62.5	0	0	0	20	0	30.5
	364	85	70	72.5	0	0	3.5	26.4	O	41.3
W7	330	72.5	70	60	0	56	0	0	86.3	0
	386	75	92.5	57.5	0	67.5	0	0	91.9	0
1/6	115	77.5	67.5	72.5	0	W.	0	O	W	0
	118	55	62.5	35	0	23.9	0	0	100	0
W9	110	77.5	00	65	0	3.1	0	0	93.6	0
	317	95	95	87.5	0	0	0	0	84.2	0
W10	254	85	67.5	72.5	0	0	0	11.8	22.1	6.9
	322	75	75	72.5	0	0	0	13.3	10	20.6
WMS	Str.I	87.5	92.5	07.5	0	65	11.4	17.1	91.9	20
MMS	Str.II	72.5	62.5	72.5	24	0	24	100	0	100
CI	**	85	92.5	87.5	0	O	0	2.9	0	2.9
C2	**	90	02.5	75	0	0	0	0	0	0
СЗ	**	87.5	82.5	100	0	0	0	0	9.1	0 .
C4	**	92.5	92.5	62.5	0	0	3	0	0	3
C5	**	97.5	90	67.5	0	2.0	0	0	16.9	0
C6	**	77.5	95	97.5	0	0	0	0	0	0
C <b>7</b>	**	95	90	97.5	0	0	0	0	0	0

TABLE V: Pathogenicity of 16 Muskmelon Isolates and 17
Watermelon Isolates.

STRA-	ISOLATE	% GI	ERM INAT	TION	% DI	SEASED	6 DAYS	% [	ISEASE	D 18 DAYS
IN		В	P	I	В	P	I	В	P	I
M2	300	90	92.5	75	20	0	53.4	83.5	5.4	96.7
	382	82.5	75	67.5	27.2	0	40.6	90.9	0	96.2
M3	88	62.5	60	65	28	0	69.3	92	0	100
	240	65	60	90	7.7	0	0	19.2	29.1	5.5
M4	162	50	72.5	87.5	10	0	8.8	80	0	51.4
	224	60	75	67.5	50	3.3	85.2	87.5	0	100
M5	151	60	50	52.5	8.4	0	38	91.6	0	100
	130	85	80	85	0	0	0	0	0	0
M6	149	67.5	40	80	0	0	0	0	0	0
	122	75	72.5	72.5	0	3.4	3.4	13.3	3.4	13.5
M7	359	90	70	75	0	0	0	0	0	0
	384	72.5	85	62.5	0	0	10.6	3.4	0	12
M8	357	82.5	80	87.5	0	0	0	0	0	2.8
	74	75	87.5	80	0	0	0	10	2.8	6.2
м9	93	85	92.5	75	8.8	0	3.3	41.1	0	26
M10	208	90	82.5	92.5	19.4	0	56.8	75	0	75.6
W2	316	92.5	90	95	0	25	0	0	86.4	0
	230	72.5	80	82.5	0	25	0	0	87.5	0
W3	167	85	87.5	75	0	0	0	0	0	0
	367	87.5	85	87.5	0	0	0	0	0	0
W4	353	92.5.	75	62.5	0	0	0	8.1	3.3	9.1
	381	.75	77.5	92.5	0	0	0	10	0	8.1
<b>N</b> 5	346	85	85	90	0	0	0	0	0	0
	380	92.5	85	97.5	0	0	0	0	0	0

TABLE V: (CONTINUED)

STRA	ISOLATE	, ,	(GERM)	NATION	% 1	ISEASI	E <b>D 6</b> DA	YS % E	ISEASE	D 18 DAYS
-IN		В	P	1	В	P	1	В	P	1
W6	217	90	80	80	27.8	0	6.3	27.7	0	46.8
	362	75	82.5	85	6.6	0	5.9	26.6	3	41.1
W7	304	77.5	75	70	0	30	0	0	100	0
	323	87.5	85	87.5	0	20.5	0	0	88.2	0
W8	117	70	67.5	77.5	0	33.3	3.2	39.2	85.1	6.4
	118	87.5	42.5	55	0	23.5	O	0	100	0
W9	116	87.5	90	95	0	22.2	0	0	100	0
	112	77.5	90	95	0	20	0	0	94.4	0
MIO	202	52.5	90	87.5	19	5.6	2.8	23.6	11.1	11.4
WMS	Str.I	82.5	75	87.5	0	33.3	0	O	100	0
MMS	Str.I	52.5	57.5	90	47.6	0	36.1	100	0	100
Cl	**	75	75	75	0	0	0	0	0	0
C2		90	95	100	0	0	0	0	0	0
C3		75	80	100	0	0	0	0	0	0
C4	•	50	70	90	0	0	0	0	0	o
C5		100	100	100	0	0	0	5	0	2.5
C6	**	70	75	75	0	0	0	0	0	0
C7		90	95	100	0	0	0	0	0	0

TABLE VI: Pathogenicity of 19 Muskmelon Isolates and 13 Watermelon Isolates.

STRA	ISOLATE	% 6	ERMINA	TION	% dis	EASED	6 DAYS	% DI	SEASED	18 DAYS
-IN		В	P	1	В	•	1	В	P	I
N2	281	60	85	62.5	21.0	0	36.4	87.5	2.9	71.8
	295	75	77.5	75	16.6	0	13.3	73.3	0	06.6
	290	70	77.5	67.5	25	16.1	20.6	62.1	16.1	82.8
	276	90	60	87.5	0	0	2.9	60.5	0	85.7
мз	123	77.5	77.5	05	3.2	6.4	14.7	36.7	45.1	64.7
83	219	U5	82.5	77.5	23.2	0	22.6	73.5	0	93.5
	277	62.5	80	67.5	15.1	6.3	25.7	67.5	12.5	85.7
	65	77.5	82.5	65	0	9.1	20.6	36.7	10.5	73.5
<b>84</b>	221	72.5	77.5	75	30	0	30	100	9.6	60
	242	82.5	75	62.5	0	0	0	67.0	0	75.7
	170	07.5	02.5	87.5	22.8	6.1	25.6	05.7	0	60.5
	231	65	65	62.5	11.7	0	9.1	79.4	2.9	60.6
115	131	77.5	72.5	77.5	0	0	0	0	0	0
	160	62.5	77.5	02.5	0	0	0	0	0	0
86	144	72.5	77.5	65	20.6	3.2	30.0	06.2	19.3	92.3
	67	60	67.5	62.5	0	0	0	0	0	0
M7		75	60	75	()	0	0	0	0	0
	383	72.5	67.5	60	0	O	0	0	0	0
NU	66	90	85	62.5	6.4	0	14.1	31.2	0	69.4
	300	02.5	77.5	72.5	0	40.6	0	0	92.1	
W2	345	67.5	87.5	85	0	30	3.1	6.5	87.1	8.6
	237	62.5	90	05	0	71.4	0	0	100	0

TABLE VI: (CONTINUED)

STRA	ISOLATE	% GF	RMINAT	NOI	% 1	DISEASE	D 6 DA	rs %	DISEAS	SED 18 DAYS
-IN		В	P	I	В	P	1	8	P	1
W3	178	80	77.5	50	0	0	0	0	0	0
	163	85	77.5	62.5	0	0	0	0	0	0
	238	90	75	75	4	29	6.3	12.1	80.3	9.3
W4	349	62.5	72.5	70	0	0	0	0	0	0
W5	342	67.5	70	67.5	0	O	0	0	0	0
	332	77.5	87.5	82.5	0	0	0	0	0	0
W6	319	90	87.5	90	4.1	0	8	28.6	0	50
	378	85	85	65	7.1	2.7	18.6	37.4	9.3	18.6
W7	158	95	100	100	0	62.1	3	6.4	100	12.1
	347	52.5	57.5	67.5	0	65.1	4	8.5	100	4.
WMS	Str.I	52.5	80	75	0	39.6	0	0	86.1	O
HHS	Str.I	100	82.5	72.5	14	0	21	89.1	7.3	90.6
Cl	•	85	77.5	75	0	0	0	0	0	0
C2	**	72.5	87.5	65	0	0	0	0	0	0
C3	*	77.5	62.5	92.5	0	0	0	0	0	0
C4	**	75	30	72.5	6	0	3.4	14.0	0	6.0
C5	**	95	75	60	12.1	16.1	5.1	63.3	75	50.4
C6	"	72.5	70	67.5	0	0	0	0	0	0
C7	11	82.5	90	92.5	0	0	0	0	0	0

TABLE VII: Pathogenicity of 18 Muskmelon Isolates and 18 Watermelon Isolates.

STRA	ISOLATE	% GERM	INATIO	N	% DISEAS	ED 6 E	MYS	% DISEASED 18 DAYS			
-IN		В	P	1	В	P	1	B	P	1	
,M2	298	72.5	82.6	72.4	45.1	0	26.0	75.1	3.1	78.3	
	260	65	62.6	82.5	51.3	0	56.4	92.1	0	100	
мз	195	50	81.3	80	50.	0	66.6	100	0	100	
	176	85	90	70	0	0	0	4.1	0	0	
M4	194	90	67.5	77.5	0	0	0	55	0	54.1	
	215	97.5	82.6	80	0	0	12.5	92.5	9.1	67.5	
M5	147	60	85	87.5	O	0	0	0	0	0	
	375	76	97.5	80	0	0	0	0	0	0	
M6	256	65	90.3	82.6	0	0	0	0	O	0	
	355	04.1	88.1	90	0	0	0	0	0	0	
117	354	82.5	75	90	0	0	0	0	0	0	
	374	75	77.5	84.1	0	0	0	0	0	0	
MO	84	80	62.6	50	6.3	0	10	6	0	10	
	283	77.5	80.1	76	3.2	3.3	0	16.1	3.3	0	
M9	98	60	95	65	0	0	0	54.1	5.3	38.2	
	127	87.5	85	60	11.4	0	13.3	37.1	0	16.7	
MIO	135	95	60	77.5	23.6	0	12.9	79	0	80.6	
	264	77.5	77.5	60	9.7	0	21.8	74.1	0	78.1	
w2	336	87.5	82.5	77.5	0	39.4	0	5.1	62	14.3	
	385	62.5	72.5	87.5	0	50.1	O	0	93.6	0	
113	173	100	72.5	85 ,	0	0	0	0	0	0	
	382	80 .	87.5	75	0	0	0	0	0	0	
W4	352	75	80	67.5	0	0	0	50	33.4	45.6	
	365	62.5	90	62.5	12.1	0	0	18.3	30	52.1	

TABLE VII: (Continued).

STRA	ISOLATE	% GE	RMINAT	TON	% DIS	EASED	6 DAYS	% DISEASED 18 DAYS		
-IN		B	P	I	В	p	1	В	P	I
W5	159	97.7	75	95	0	0	0	0	0	0.
	216	82.5	72.5	97.5	0	0	0	0	0	0
W6	168	85	87.5	00	0	0	0	23.2	0	31.2
	364	70	75	85	0	0	0	25	0	47
167	330	72.5	77.5	80	ō	35.4	0	0	80.8	0
	386	92.5	72.5	75	0	55.2	0	0	72.5	0
WO	115	75	75	72.5	0	N	0	0	W	0
	118	80	77.5	87.5	0	19.3	0	0	83.8	0
W9	110	77.5	85	80	0	18.7	11.5	0	81.1	15.0
	317	95	95	80	2.6	2.6	0	7.9	86.9	0
W10	254	87.5	75	75	0	0	0	0	0	0
	322	40	80	80	0	0	0	0	0	0
WAIS	Str.I	60	75.	72.5	6.3	63.4	0	15.6	90	0
MNS	Str.II	75	80	72.5	20	6.3	0	90	15.6	0
Cl	**	65	87.5	72.5	0	0	0	0	0	0
C2	•	82.6	84.1	75	0	0	0	0	0	0
сэ		77.5	75	80	0	0	0	0	0	0
CA		92.5	80	75	0	0	0	0	0	0
C5	**	72.5	72.5	80	0	0	0	0	0	0
C6	**	62.5	50	65	0	0	0	O	0	0
<b>C7</b>	**	82.6	75	72.5	0	0	0	0	0	0

considering the 108 combinations of host and pathogen (36 Bender's, 36 Peerless, and 36 Iroquois with each Fusarium strain), 89 of the results for both experiments did not vary more than 10% from one experiment to the other. If the allowable variation were increased to 20%, then 99 of the results would be similar for both trials. Because of the similarity of results the writer feels that the pathogenicity of the isolates has been compared in a reliable manner.

Tables IV to VII show that pathogenicity is correlated with the morphological types previously designated. Considering the M2 class of Tables IV, V, and VI we find that the degree of pathogenicity is approximately the same, except in two cases. M2 Table IV isolate 298, Iroquois seedlings and M2 Table VI isolate 290, Peerless seedlings. If one studies the rest of Tables IV, V, and VI the close agreement of results within a given morphological type can be found in 173 of the 201 muskmelon isolates. Certain morphological types might have slight pathogenicity variation accounting for the difference in results.

This experiment also showed the attack of watermelon seedlings by muskmelon <u>Fusarium</u> and vice versa. For example see Table V, isolates 300, 240, and 122. Other examples can also be noted.

Results also indicate variation in strain type, and pathogenicity from individual farms. Isolate 300 from the Crouchley farm in Aldershot placed in strain 2, gave 83.5% wilt with Bender's, 5.4 with Peerless, and 96.7% with Iroquois. Isolate 264 from the same farm gave 76.7% wilt with Benders, 0% with Peerless, and 77.4% with Iroquois. Isolate 122 from Crouchley farm, placed in strain 6 gave 13.3% with Bender's, 3.4% with Peerless, and 13.5% with Iroquois. Further examples of this can be noted

with isolates from various other farms.

3. Investigations on the Extent to Which the Pathogens Can Attack Both
Hosts and Whether One of Them Could, by Natural or Induced Mutation
Give Rise to the Other.

If such mutations occur naturally, then the problem is, how could such a mutation be detected? If large numbers of conidia could be inoculated into immune host plants, and the host plant became diseased, then if appropriate controls were used it could be assumed a mutation had occured, conferring on the pathogen the ability to attack a previously immune host. If this mutation does occur then its frequency would be increased by treatment with such mutagenic agents as ultra-violet light, X-rays, and such chemical agents as the methyl xanthine derivatives.

It was felt that the results of the pathogenicity trials previous—
ly reported, indicated that certain isolates had the ability to attack
both hosts. To confirm this, a further pathogenicity experiment was carried
out. Selected strains were inoculated into 17 flats of sterilized soil.
Sixty seeds of muskmelon (Bender's Surprise), and 60 watermelon (Peerless)
were planted in each flat, and disease incidence was estimated by observing
the emergence and wilt that followed. The results are listed in Table VIII.

Considering the muskmelon isolates first, it is evident that pathogenicity varies from strain to strain. This is in accord with the results of the pathogenicity trials previously reported. Severity of attack varied from very severe to little or no effect on the muskmelons, while certain strains also attacked watermelons (strains 2, 3, 9, and 10), but much less severely.

Considering the watermelon strains it was found that the range was

TABLE VIII: Comparison of Pathogenicity of Isolates from Muskmelon and Watermelon to Both Hosts.

STRAIN		MUSKME LON							WATERMELON					
		EMERGENCE			POS!	POST-EMERGENCE			EMERGENCE			POST-EMERGENCE		
		%		MOR	MORTALITY IN			%			MORTALITY 1			
					29	DAYS	%				29	DAYS	%	
		T 1	T 2	T 3	T 1	T 2	T 3	T 1	T 2	<b>T</b> 3	T 1	T 2	T 3	
1.1	iuskmelon	83*	87*	73*	62	89	86	814	100%	92#	0	0	0	
2.		72	83	65	100	95	87	68	80	75	6	8	0 *	
3.		48	50	47	93	90	81	78	78	87	14	27	20	
4.		70	73	81	48	51	44	82	51	<b>01</b>	0	0	0	
5.	**	87	90	85	0	0	0	72	78	85	0	0	0	
6.	*	81	58.3	82	0	0	0	50	82	75	0	0	0	
7.	**	73	80	52	0	0	0	95	88	92	0	0	0	
8.	**	68	73	67	35	39	27	72	95	62	0	0	0	
9.	**	85	100	95	48	56	50	76	83	83	41	42	56	
10.		82	78	92	85	83	90	85	82	92	15	17	10	
		* R	eplic	ates	#	Rep1	icates							

T Denotes Trial

TABLE VIII: (CONTINUED).

STRAIN				MUSH	MELON			WATERMELON						
			EMERGENCE			POST	-EMERG	ENCE	EN	ERGEN	CE	POST-EMERGENCI		
			%			MORTALITY		IN		%		MORT	IN	
						29	DAYS	%				29	DAYS	%
	1.Wate	ermelon	90	83	87	16	12	11	82	78	87	100	96	100
	2.	"	90	75	78	10	6	9	78	87	82	93	91	82
	3.		68	78	83	0	0	0	52	63	58	3	5	0
	4.	**	82	81	85	42	51	56	85	65	70	20	26	29
	5.	**	73	80	83	0	0	0	88	92	80	0	0	0
	6.		75	60	92	0	0	0	100	90	83	0	0	0
	7.	99	82	78	43	0	0	0	82	51	62	81	93	87
	8.	•	48	52	78	0	0	0	85	100	87	100	100	92
	9.	*	78	82 /	85	0	0	0	90	100	82	84	71	36
	10.	**	90	100	65	13	17	25	81	57	85	0	0	10
	Check	1	90	83	78	0	0	0	100	92	85	0	0	0
	Check	2	68	100	92	0	0	0	82	85	83	0	0	0
	Check	3	50	81	87	0	0	0	65	80	92	0	0	0

was from complete pathogenicity, to non-pathogenic forms. Strains were also found (1, 2, 4, and 10) which attacked muskmelons with less virulence than watermelons.

It is an interesting point that both the isolates from muskmelon and watermelon include strains which cannot attack either host. Certain isolates from watermelon (1, 2, 3, 7, 8, and 9) caused severe wilting. This is in accord with the results of McKeen (13). Strains 3 and 4 of watermelon caused a mild attack and strains 5 and 6 showed no effect. In a preliminary trial using 4 watermelon strains (1, 3, 3\*, and 10) none of the isolates attacked watermelon.

The results obtained with the muskmelon isolates were similar, ranging from severe pathogenicity to non-pathogenic.

A point of interest is that during field collections, 5 isolates of a cucumber <u>Fusarium</u> were obtained. These were not included in these trials, but in a preliminary test it was found that the cucumber <u>Fusarium</u> appeared to attack both host types of melon.

In no instance did an isolate from one host plant, attack the other host plant with the same virulence as it did its original host.

### 4. Some Experiments on the Biology of the Pathogens.

(A) Investigations into the Possible Occurrence of Meterothalism Among the Collected Strains of <u>Fusarium</u>.

The purpose was to determine whether by pairing representative isolates the perfect stage of the pathogens could be obtained.

In each experiment ten select strains were paired in all combinations by mixing prepared spore suspensions, which had been adjusted by means of a haemocytometer to approximately 1000 conidia per ml. Ten sets of ten isolates were studied in this manner.

TABLE IX: Results of Pairing a Number of Isolates from Muskmelon in All Combinations.

	MMS	128	88	224	130	355	384	74	98	135
MMS		-	*	•		•	•	ng kanan <del>an</del>	-	•
128	-	-	-	•	•	-	•		+	
88	•	•		-	•	iate	-	-		•
224	•	•	-	•	•	•	-	-		-
130		•	-	+	+	•	-	-	-	+
355	-	-	•	•	•	-	•	-	•	-
74	-	•	-	-	-	-	-	-	-	•
98		+		+		•	-	•	-	•
135	•	•	-	**	+	+	-	+	+	+

+ Denotes presence of small dark dots, on or in the agar of the plate.

TABLE X: Results of Pairing a Number of Isolates from Watermelon in All Combinations.

	WMS	385	367	381	230	364	330	115	317	304
WMS	+	+	+	+	+	*	-	-	-	-
385	+	-		•	•	•	•	-	•	•
381	•		+		•	•	•	-	-	-
230	+		+	•	+	+	-	-	-	
364	+	+	+	+	•	-	•	•	-	-
330	-	-	-	-	-	-	4	•		-
115	-	-	-	-	-		-	-	-	-
317	•	-	•		1.	-	•	•	•	-
304	•	-		-	-	•	-	-		

TABLE XI: Results of Pairing a Number of Isolates from Muskmelon in All Combinations.

	MMS	281	277	170	131	160	87	358	383	242	229
MMS	-	•	•	+		•	•	•	+	•	+
281	•	+		+	•		•	+	+	-	-
277	• 1		•	+		•	*		*	•	•
170	+	+	+	+	•	+	*	+	•	-	+
131		*	•	•	-	-		•	•	*	-
160	•	•	-	•	-	+	•	•	+	-	-
87	-	•	-	•		•	*	-			*
358	•	+	•	•	-	-	-	-		•	+
383	+	+	•	+	-	•		-	-	•	+
242			-	+	-	-	-	+	-		-
229	+	+	+	+	•			+	+		+

TABLE XII: Results of Pairing a Number of Isolates from Muskmelons and Watermelon in All Combinations.

	MMS	281	277	170	131	160	87	358	383	242	
WMS		•		•	+	•		•	•		
385		+				•	+ .	•	•	-	
367			•	-	•		-	-			
381	-	•		-	-	-	•	- 7	•	-	
230		+	+	+	-	-	-	-		-	
364	•		•	•	+	4	1	•	-	-	
330		•	+	+	-	-	+	•	-	-	
115	•	-			+		-	•			
317	-	+	+	+	•		•	+	**	•	
304	•	-	•	-	•		•	11.	-	-	

The plates were poured with 15 mls. of Potato Dextrose Agar + 1%
Yeast Extract, into which was mixed 1 ml. of spore suspension of both
strains, which were to be paired.

A total of 100 such pairings were carried out, but due to results only 40 are reported here and are listed in Tables IX, X, XI, and XII.

Dark hard structures were noted throughout the plates, and their identity investigated. It was thought that they might possibly be perithecial primordia. Strips of agar containing the structures were soaked in 95% alcohol and thin sections were made. Although on repeated occasions they were found to be hollow masses of mycelia and swollen cells, no evidence of asci or ascospores could be found.

(B) The Effect of Exposing Conidia of Muskmelon and Watermelon

Fusarium to Various Mutagenic Agents.

The original purpose of this experiment was to determine the dosage of ultra-violet light that would cause about 50% mortality in conidial suspensions. It was considered that the mutation rate of conidia so treated would be the maximum possible without too great a loss of viability. The treated conidia were to be injected into both hosts. By observing the injected plants it could thus be determined whether mutant types had arisen capable of attacking the other host.

Preliminary experiments indicated that neither host was immune to injected non-treated conidia of the other pathogen, but other effects were found.

(i) The effect of ultra-violet light.

Strain 1 of the muskmelon <u>Fusarium</u> and strain 1 of the watermelon <u>Fusarium</u> were the two strains selected for these experiments.

For treatment of the conidial suspensions with ultra-violet light, the selected strains were grown on Potato Dextrose Agar in petrie dishes and conidial suspensions were prepared by flooding the plates with sterile water. With a haemocytometer the suspensions were adjusted to contain 2000 conidia per ml. One hundred mls. of this suspension were added to a moist chamber dish 5 inches in diameter to which a layer of water agar had been added to make the bottom perfectly flat. Then the suspension was irradiated, being gently agitated with a rocking motion all the while, with a Westinghouse Sterilamp (WL-782-20) at a distance of 16 inches. The irradiation was interrupted at the proper times to allow withdrawal of 1/10 ml. samples to be plated. Plating was done in Czapek Solution agar + 1% Oxgall to delimit the colonies, as described by Miller et al. (17), and a similar set plated into Czapek Solution agar without Oxgall for comparison.

The results of the experiment are listed in Table XIII.

It is evident that periods of irradiation up to 1 minute did not affect the viability of the conidia, but after 2 minutes decrease was noted, and this was pronounced after 5 minutes. Only one colony was found in the plates exposed for 15 and 30 minute periods.

On the Oxgall plates, the colonies did not intermingle and mutations could be detected more easily. They were made up of four main types:

- (1) Those with a "stringy" mycelium, which were light lime in colour,
- (2) a fine cottony colony with a periphery marked by a fine band of mycelia.

  Light lime in colour, (3) a rough, white, rather spreading colony. The

  mycelia of which is semi-appressed, (4) blue tinted colonies of various

  forms.

The plates which did not have Oxgall added, consisted of one large

TABLE XIII: Effect of Ultra-violet Light on Germination Conidia

of the Muskmelon Wilt Fusarium and Watermelon Wilt Fusarium.

TIME OF						
EXPOSURE TO		RUSKRELL	N		WATE I	RELON
ULTRA-VIOLET	NUMBER	OF COLOR	IES THAT	NUMBE	K OF C	COLONIES THAT
LIGHT	www	PED PER	PLATE	DEV	LOPED	PAR PLATE
O Sec.	100 **.	116 *.	117 *	123,	136,	127
10 Sec.	95,	68.	97	135,	133,	122
30 Sec. V	102.	107.	181	126,	128.	130
l Min.	98.	98.	105	100,	131.	120
2 min.	46.	55.	50	73,	70.	69
5 Min.	12.	7.	14	21,	14.	16
15 Min.	0.	1.	0	0.	0.	0
30 %in.	0.	0.	0	0.	0.	0

<sup>\*</sup> Replicates.

colony resulting from the intermingling of numerous colonies. The colonies varied in texture and appearance from place to place, and detailed examination was made for evidence of the perfect stage as a result of this treatment. Several types of abnormal structures were noted, but repeated sectioning gave no indication of perithecial development.

It is evident from the table, that plating of 1/10 ml. samples of suspension adjusted to 2000 conidia per ml. did not give rise to the expected 200 colonies per plate. This could be explained by: (1) not all the conidia in the suspension were viable and (2) during preparation of the concentrated suspension, the suspension had to be centrifuged. This might have resulted in clumping of cells, thus reducing the actual number of colonies that would be produced. Either one or a combination of both these factors might have been responsible.

(ii) The effect of Caffeine.

Fries (4) first reported success with methyl xanthine derivatives as mutagenic agents.

A conidial suspension was prepared in the manner previously mentioned using the muskmelon standard strain and the watermelon standard strain. The strength of the suspension was not adjusted to any predetermined value since it was felt a maximum number of conidia should be subjected to the treatment, and the dilutions utilized to get a reduction in numbers.

The suspension density for muskmelon was 220,000 viable conidia per ml., and 300,000 viable conidia per ml. for watermelon. The Caffeine was added to the stock suspensions in an amount sufficient to make the final Caffeine strength 0.2%, a figure determined to be adequate by Fries (4). The suspensions were placed in the incubator, samples were taken on 6 occasions, and plated into Potato Dextrose Agar + 1% Yeast Extract.

In performing the sampling the stock suspension was thoroughly shaken, then 10 mls. were removed, and placed in a dilution bottle containing 90 mls. of sterilized distilled water. Ten 1 ml. samples were plated from this bottle, and a 1 ml. sample was also removed and placed in a dilution bottle containing 99 C.C. of sterilized distilled water. From this 10 one ml. samples were removed and plated. The plates were then placed in the incubator and examined periodically.

All plates were allowed to remain in the incubator, then at intervals of 1, 2, and 3 weeks they were examined. In no case was there any clear example of perithecial development. Many of the hard black structures previously mentioned were apparent, the number increasing up to the 72 hour count, none being apparent after that. This probably indicates these structures are the result of the mutagenic treatment.

The results are shown in Table XIV. The types of colonies produced included all four types produced by ultra-violet irradiation, plus a creamy coloured colony with a somewhat appressed mycelium.

TABLE XIV: Effect of Caffeine on Germination of Conidia of the Susknelon and Sater-

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MINNELLON

		MUNICIPAL OF COLORIES	NEED OF GOVIES	2183
	COUNT I (ISL.DAY) COUNT 2 (2nd.DAY)	COUNT 2 (Sad.nav)	COUNT I (1st. DAY) COUNT 2 (2nd.DAY)	COUNT 2 (2nd.DAY)
12 115.	* 1.3	* 9.69	* 0.72	*1.17
24 hrs.	23.23	20.03	27.4	29.6
of hrs.	25.2	24.6	26.4	57
72 hrs.	17.4	17.6	16.4	17.6
% hrs.	•	**	9.11	**
120 hrs.	2	**	-	**

\* Average of ten counts.

\*\* Colonies overgrown, unable to count.

(iii) The effect of Theophylline.

Theophylline, another methyl xanthine derivative, was also tested using the same technique as with Caffeine.

The density of viable conidia in the suspensions employed was 295,000 conidia per ml., for the muskmelon <u>Fusarium</u> and 319,000 conidia per ml., for the watermelon Fusarium.

at the intervals mentioned in the previous experiment were of the wild type in nature, and three of the variant colony types previously mentioned. The rough, white, rather spreading colony with the semi-appressed mycelia was absent. The creamy coloured type of colony which was first noted with the Caffeine, was also present here. The hard dark structures were present after the 3-week period, as well as local areas of mounded dark brown mycelia. These mounds were filled with numerous macrospores. On single spore isolation of one of these spores, a mutant was found which produced only large macrospores, and these were produced in rings. This phenomenon had been noted earlier with other organisms, and had sometimes been found to be the result of light striking the cultures.

Plates of Czapek agar were inoculated in the centre, wrapped in photographic paper, and sets incubated at 50°C., 37°C., 27°C., and one set placed in the refrigerator. These were examined after 6 days, but no ring was apparent and sporulation was uniform throughout the plate. Light was next tested as the cause of this ring effect in sporulation.

Plates were inoculated with the mutant, wrapped in black photographic paper, and incubated at 27°C. These plates were exposed to light from the north window of the laboratory after 2 days incubation. Six plates were

TABLE XV: Effect of Theophylline on Germination of Conidia of the Muskwelon and Watermelon

ill Fusaria.	
	Total Control
	The state of the state of
*90	
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		RINKE OF CLOSIES	NAMES OF COLMES	SHES
		(1st. Day) Court 2 (2nd.Day)	COUNT ALSEA	COUNT MIST. DAY) COUNT 2 (2nd.DAY)
12 113:	* 0. 737	124.4 *	126 *	* 011
24 hrs.	3	ŝ	3	8
46 hrs.	7	9	7	25
72 hrs.	12	14		12
96 hrs.	4	**	•	*
120 hrs.	0	**	0	**
	STORY *	Average of ten counts.		

\*\* Colonies overgrown, unable to count.

removed from the light after 0, 5, 10, 15, 30 minutes, 1 hour, and 2 hours. The peripheral edge of each colony was marked on the underside of each plate, at the time of exposure. Upon removal from the light the plates were immediately wrapped again and returned to the incubator. The light intensity falling on the plates was 26.5 foot candles per square foot throughout the entire period, measured with a General Electric Light Meter.

On examination after 3 more days, it was found that the plates not exposed to light, did not have the spores produced in a ring, but were randomly scattered throughout. Every plate from the 5 minute time up to the 2 hour time contained a ringing effect in which the organism produced its spores. The ring effect was produced directly over the marked line, indicating it was exposure to light which caused the ring type sporulation.

(iv) The effect of visible light on cultures of <u>Fusarium</u> irradiated with ultra-violet light.

In the preceeding ultra-violet irradiation experiment the plates containing the irradiated conidia were immediately placed in the dark. The advisability of this follows from the discovery of Kelner (9) that visible light to some extent reverses the physiological effect of ultra-violet light. Not only was the survival of irradiated cells of Escherichia coli increased by subsequent exposure to visible light, but the numbers of mutations were reduced. Light induced recovery was noted with Escherichia coli. Streptomyces griseus. Penicillium notatum, and Saccharomyces cerevisiae.

An experiment was performed to determine whether a similar effect could be detected with <u>Fusarium</u>. Thirty-two plates containing 15 ml.

Czapek Solution agar were inoculated in the centre with strain 1 of the muskmelon and 32 plates with the watermelon pathogens. After the colonies

had grown so as to half cover the plates they were removed from the incubator (27°C.) and irradiated as before in lots of 4 for each organism for
periods of 0, 10, and 30 secs., 1, 2, 5, 15, and 30 minutes. After each
irradiation 2 plates of both organisms were placed in the dark in the
incubator and the other 2 plates were exposed to the light of three 100watt desk lamps for 6 hours and then placed in the incubator.

A week after treatment the plates showed no apparent difference between the "dark" and "light" after-treatments with the watermelon Fusarium. With the muskmelon strain, however, a distinct difference was noted in the plates exposed for 2 and 5 mins. Those kept in bright light for 6 hours did not develop the bluish ring produced in the darkened plates. The plates exposed for less than 2 minutes developed no ring in the light or dark, and those exposed longer than 5 mins. developed it in both light and dark.

Thus visible light evidently nullified an effect of exposure to ultra-violet light.

(v) The effect of x-rays on cultures of Fusarium.

It was found impossible to irradiate spore suspensions with x-rays as was done with the ultra-violet light, because of the nature of the apparatus available.

Plates of Czapek Solution agar were inoculated in the centre with the muskmelon and watermelon standard strains. Instead of placing the petrie dish lid on, the bottom half was wrapped with sterilized paper and sealed. After 3 days incubation the plates were exposed to x-rays of the machine in the Physics Department, McMaster University.

The time of exposure was 0, 5, 10, 15, and 30 seconds, 1, 5, and 15 minutes.

After irradiation through the paper cover, the paper was removed and replaced by the glass lid.

The plates were examined at the designated intervals of 3 days for a four week period but no effect of x-rays was noted.

# 5. Assimilation by Certain <u>Fusarium</u> Isolates of Various Carbon Sources for Growth.

Ten isolates were selected one from each of the chief morphological types. Single spore isolations of each were made by the method described by Keitt (8). Stock cultures were maintained on Potato Dextrose Agar + 1% Yeast Extract slants, covered with sterilized Paraffin Oil, then stores in the refrigerator.

The cultures selected were: muskmelon standard, watermelon standard, isolate 88 (muskmelon strain 3), isolate 74 (muskmelon strain 8), isolate 355 (muskmelon strain 6), isolate 130 (muskmelon strain 5), isolate 330 (watermelon strain 7), isolate 385 (watermelon strain 2), isolate 230 (watermelon strain 2), and 364 (watermelon strain 6).

The medium selected for this experiment was Wickerham's Yeast

Nitrogen base (a carbon-free medium) which has been used successfully by

Wickerham (24) in studying the utilization of various carbon sources by

yeasts.

Into carbon-free medium, inoculum from each stock tube was placed.

This was allowed to grow for four days, and from each of these tubes inoculum was placed into more tubes of carbon-free medium. It was from this second set of carbon-free medium tubes, that inoculum was taken for the experiment. This procedure insured that little or no carbon source would

be carried over in the inoculum.

The compounds to be tested as carbon sources were added in 1% strength to the medium.

Sugars tested were:-

### Monosaccharides:

Bexoses: Fructose, Glucose, Galactose, and Mannose.

Pentoses: L-Arabinose, d-Xylose, and d-Ribose.

#### Disaccharides:

Maltose, Cellibiose, Lactose, Melibiose, and Saccharose.

## Trisaccharides:

Raffinose.

## Polysaccharides:

Araban, Starch, and Inulin.

Other substances tested were: - pectin, sodium acetate, malonic acid, succinic acid, tyrosine, glutaric acid, tryptophane, methionine, histidine, alanine, acetic acid, glycerol, peptone, yeast extract, sodium pyruvate, sodium malonate, mannitol, sodium propionate, sodium butyrate, sodium formate, and glycine.

Each tube was adjusted to pH 7. Control tubes were employed in which no carbon source was added.

In Table XVI is recorded the amount of growth and type of sporulation for each. The degree of growth for each isolate was assessed by comparison of each tube against a set of standards prepared and kept under oil.

It is interesting to note that in a few cases certain compounds can be utilized by some of the isolates, but not by others. An example of this is isolate 355 when Histidine is used as the test source. Frequently certain

TABLE XVI: Growth and Sporulation of Selected Isolates from Muskmelon and Watermelon on Various Carbon Sources.

## ISOLATE.

CARBON SOURCE	MAS	MMS	88	330	305	74	355	230	130	364
Waltose	2½*; m	2:Nm	2;(M)m	2½;(M)m	2;Mm	3;Mm	2½(M)(m)	2;(質)(由)	2;(M)m	1;
Lactose	及。(M)(m)	从;(M)(m)	为;Mm	1/2: Min	½;Mm	从(图)(m)	<b>%</b> ; a	1;Nm	1;Mm	2:N
D-Galactose	3;8/m	3;Mm	1½;Mm	2;m	2;No	2;Mm	1½; Mm	2;Mm	2½; Nm	1;9m
Fructose	3;(屬)(丽)	2;(屬)(m)	3;(M)	2½;(圖)(m	) 3;(M)(n)	)3;m	3;(新)(m)	2½; Mm	2½:n	<b>%</b> ;
Melibiose	31/2; Mm	2:0	2½;(m)	2½;m	2½;m	2½;m	2½;(M)(m)	31/2; 8km	2½;m	1;m
Saccharose	3;(%)m	2;(M)m	2;	25;m	3;(M)m	2½;n	2½;m	3;(%)(m)	3;m	1/21
Dextrose	1½;(屬)m	25;(图)m	2;(M)m	4;(M)m	2½; Mm	4;(%)m	21/2:	34; Mm	3;m	<b>%</b> :
D-Nannose	3;(質)m	2;(N)(m)	2;(M)(m)	)3;m	25; Mm	3;m	2;m	3/2:0	2½:陽冊	1;8
Arabinose	3;(m)	2;(m)	2;½(m)	2½;m	2½;(層)(m	)3½; (N)m	2½;m	3;Mm	2½;(图)面	经;简
Raffinose	3;(M)m	2/2;	2½;m	4;m	2;Nm	3/2;Mm	3;m	2½;(剂)m	3)2; (N)m	1;
Inulin	3/2; Mm	2;简(图)	3;(m)	2;(M)m	2;Mm	2;(M)m	2;Mm	2½; Nim	2½; Ma	2;M(m)
0-nylose	3;m	2;(a)	3;	3;(H)m	2版;(M)m	3½;(M)m	2;m	34;m	3%;(M)m	层;M(m)
Succinic Acid	2;M(m)	1½;m	2;(m)	2;(m)	2;m	2;m	2;m		1岁;(丽)	i
Histidine	1; Min	1%;(图)和	1½;m	1;Ma	1;Ma	1/2;Mm	1 -1	1/2; (M)	1%; (質)m	1½; (M)m
Cellibiose	3½; Mm	3;Mm	3;	3;m	1;Mm	3;m	4;(M)m	4;m	3;N(m)	1;8
Sodium Acetat	e2%;UG	2:4	21/2;	1/2:	2;Mm	2;Mm	2½; Ma	1;	i	24;
Starch	2½:Mm	2: 4	2:Mm	1½:Mm	2:Ma	2; m	2:Mm	2;Mm	2;Mm	1:

TABLE XVI: (CONTINUED)

## ISOLATE

CARBON SOURCE	WMS	MMS	88	330	385	74	355	230	130	364
Glucose-1-										
Phosphate	3;M(m)	3;Mm	3½;m	3/2; Mm	3; Ma	4; Ma	3;	4; Mm	3;m	1;m
Fructose-1,6-										
Diphosphate	3;Nm	3; m	3½; m	3; m	3; m	3;Mm	3; m	3;Km	3; m	1; m
dlMethionine	1	•	;							;
Malonic Acid	:	•		1	•	•	•		i	
Sodium										
Malonate	1;Mm	1; m	1; m	1;(M)m	1;Mm	1; Man	1; Nim	1;Sm	1;1	1;(M)(m)
Peptone	3;Mm	2½; m	2½; n	3; m	3/2; Men	3;Mm	3;8m	3/2;M	3;Mm	3½; Max
Glycerol	3;Mm	3; m	2;	2½;(例)曲	34; Ma	21/2:18m	3;(M)m	2½; Ma	2/31m	1;(M)m
dl Alanine	3; в	3; m	3½; m	3; m	3; m	3; m	3;Ma	3; (m)	3; m	2½;(附)m
Yeast Extract	3/2; Mm	3½: n	3; m	3½; m	3;	3½;iAm	3/2; Ma	3:	3;1km	2½;稱
Sodium										
Propionate	3;Mm	3;Mm	3½; Nm	2;(附)加	3½; Mm	3;Mm	3½; Min	1; Na	3; Mm	2後;別(用)
Glycine	2;Mm	2½;m	2;(例 <b>)</b> m)	2;Mm	2;Mm	2;11m	2½; Mm	1;Ma	1½;Ma	1;Mm
Pectin	2; m	1; m	2; 8	2; m	2; 8	2; m	2; m	2; m	2; m	2; m
Cellulose	1; m	1; m								

## TABLE XVI: (CONTINUED)

- \* Average of four tubes for each test.
- 1 Slight growth.
- 2 Moderate growth.
- 3 Abundant growth.
- 4 Very abundant growth.
- M macrospores (M) or (m) present.
- m microspores but infrequent.

isolates did not sporulate, even though a fairly good degree of growth had been produced. For example isolate 88 with cellibiose and Sodium Acetate, isolate 355 with Dextrose. A great many more examples of this can be seen throughout the table.

Another point of interest is the great variation in amount of growth, and in the nature of sporulation with any one isolate, supplied with different carbon sources. For example consider isolate 230. With Succinic acid, there is no growth or sporulation. With Sodium Malonate there is light growth, producing both macrospores and microspores. With D-X ylose however, there is a very heavy mycelial growth, but only microspores are evident.

Isolate 364 which is a medium brown, appressed slightly pathogenic isolate, closely resembling many of the saprophytic <u>Fusaria</u> is especially interesting. It gave a heavy mycelial growth with only one test chemical and with a few others it gave a fairly good growth. In the majority of cases, however, it gave poor growth with little sporulation.

With one or two other isolates, the amount of mycelial growth was fairly constant, but the nature of sporulation and the amount of it varied from carbon source to carbon source. An example of this is isolate 130 which had an average growth value of between 2 and 3. Sporulation ranged from production of both macrospores and microspores to failure to produce any spores.

It should be mentioned here that the nature of the spores varied as well as the actual type of sporulation. One isolate might produce macrospores in two different test chemicals, but these spores might not be the same shape or have the same average number of cells. As an example

of this consider isolate 230. Using Dextrose as the test source, the microconidia were slightly longer than they were wide. The macroconidia were quite large, sickle-shaped, with a maximum of four cells. When Inulin was used as the test source the macroconidia were very abundant, with a great many spores being 7 celled. Most of the remaining spores were 4 celled, with a few having a smaller number of cells. These spores were extremely large, and varied from sickle-shaped types to long slender forms with little or no curvature.

## 6. Assimilation by Certain <u>Fusarium</u> Isolates of Various Nitrogen Sources for Growth.

Six isolates, each of a different morphological type, were selected for study, as follows: muskmelon standard, watermelon standard, isolate 88 (muskmelon strain 3), isolate 74 (muskmelon strain 8), isolate 230 (watermelon strain 2), and isolate 364 (watermelon strain 6). The technique for preparing the stock cultures was similar to that employed in the carbon assimilation tests.

The medium employed, as mentioned previously, was derived from Wickerham's carbon base medium (24).

Into nitrogen free medium, inoculum from each stock tube was placed.

This inoculum in the nitrogen free medium was allowed to grow for 2 days.

The growth in these tubes then served as inoculum for a second set of nitrogen free medium tubes, from which, after 2 days incubation, the inoculum for the nitrogen test tubes was obtained. This ensured that little or no nitrogen would be carried over in the mycelium.

The compound to be tested as a nitrogen source was added in 0.5% strength.

Substances tested as nitrogen sources were: - ammonium acetate, dl methionine, potassium nitrate, L (+) histidine monohydrochloride, yeast extract, dl tryptophane, asparagine, ammonium sulphate, paraminobenzoic acid, peptone, urea, alanine, ammonium nitrate, glycine, calcium pantothenate, sodium nitrate, sodium caseinate, pepsin, thiamine hydrochloride, and sodium azide.

From the results recorded in Table XVII it is hard to generalize on the type of compound that can be utilized as a source of nitrogen. For example, one cannot say that vitamins could be assimilated as a nitrogen source for thiamine hydrochloride cannot be utilized while calcium pantothenate can. It is not clear whether the calcium pantothenate is utilized as a source of nitrogen or whether the nitrogen is obtained from alanine as a result of hydrolysis of the calcium pantothenate to alanine and other compounds. This has been reported by Hawker (6) with certain other fungi. The amino acids tested generally supported growth of all strains tested, with the exception of L (+) histidine monohydrochloride which supported growth in only one isolate. Ammonia nitrogen appeared to be a ready source of nitrogen for use by most of the tested strains as does nitrate nitrogen. This is in keeping with the results found by Lilly and Barnett (12), with the exception the nitrogen from sodium nitrate is not utilized by any of the isolates.

It is evident on the basis of growth that certain test substances can be utilized by some of the isolates, but not by others. For example isolates 74, 230, and 364 could not utilize dl methionine while the other three isolates could. The muskmelon standard was the only isolate that could utilize L(+) histidine monohydrochloride, while the watermelon

TABLE XVII: Growth and Sporulation of Selected Isolates from

Muskmelon and Watermelon on Various Nitrogen Sources.

NITROGEN SOURCE	WMS	MMS	88	74	230	364
Ammonium Acetate	*	3; m	3%; (N)m	4; Mm	3;(M)m	3;M(m)
dlMethionine	1½; m	1½; m	1 ;(N)m			
Potassium Nitrate	3; m	3; m	3; m	4;(M)m	2;Mm	3;(M)(m)
L(+)Histidine						
Monohydro-chlorid	e ;	2 ;(M)m	•			
Yeast Extract	3 ; m	3; m	3; m	3;(M)m	2;Ma	3;
dlTryptophane	2½; m	2; #	2½; m	3; m	21/2; Nim	12: m
Asparagine	2; (m)	2; (m)	2; (m)		1½; (m)	<b>½:</b>
Ammonium Sulphate	1½; (m)	1%; (m)	1½; (m)	3½;(%)m	1½;(M)m	<b>%</b> :
Para-amine						
Benzoic Acid		•				
Peptone	34: 0	3; m	3; m.	4;Nm	3; Ma	3;(M)
Urea	2/4: Nm	2½; a	25; (M)m	21/4; m	2½; Mm	4:
Alanine	1%; Mm	2 ; m	2 ;(N)m	25:	2 ; m	2;
Aumonium Nitrate	2 ; m	2½; m	2½; m	2 ;(M)m	2;Mm	2;(M)(m)
Glycine	2 ;(M)(m)	2 ;(M)m	2 ; <b>(18</b> )m	2½; m	3 ; m	3; m
Calcium						
Pantothenate	1; (m)	1; (m)	1½; m	1; m	1½; Mm	4:
Sodium Nitrate	14:	<b>%</b> :	<b>%</b> ;	<b>½</b> ;	<b>%</b> :	<b>5</b>
Sodium Caseinate	2; m	14; m	1½; m	1½; m	2;Mm	1; (m)
Sodium Azide			1	•	•	
Thiamine						
Hydrochloride		16; (m)		1/2:		•
Pepsin				1½; (M)m		1½;(※)(m)

## TABLE XVII: (CONTINUED)

- \* Average of four tubes for each test.
- 1 Slight Growth.
- 2 Moderate Growth.
- 3 Abundant Growth.
- 4 Very abundant Growth.
- M macrospores (M) or (m) present
  - m microspores but infrequent.

standard was the only one which could not utilize ammonium acetate as a nitrogen source. Variation in growth was also found when asparagine, thiamine hydrochloride, and pepsin are the sources of nitrogen. This variation can range from very sparse growth, sodium nitrate with the water-melon standard isolate, to very heavy growth, yeast extract with the same isolate.

Variation in the type of sporulation occured as one changed the nitrogen source. In some cases, isolate 364 with yeast extract, alanine, calcium pantothenate and isolate 74 with alanine, there was growth without any sporulation. More pronounced, however, was the general lack of macrospores. They occured in abundance only a few times, and generally this was only with isolate 230. They were produced in abundance with other isolates on a few nitrogen sources, for example watermelon standard, using urea and alanine, isolate 74 using peptone, and isolate 364 using ammonium acetate. Microconidia were produced with the majority of the isolates in a majority of the nitrogen sources, and were generally in good numbers when present.

It was noted, however, that the actual nature of the spores in this experiment did not vary nearly as much as with the carbon utilization tests. An example of this was macrospore production with the isolate 230. The macrospores when present here were almost always large 1-7 celled, sickleshaped types.

## 7. Comparison of Resistance of Certain Selected Melon Varieties Under Field Conditions.

A field experiment was performed to compare the resistance of the varieties selected to the muskmelon <u>Fusarium</u> wilt organism. The melon varieties selected were:

## (1) Muskmelon

- (a) Bender's Surprise, susceptible, 90 day melon.
- (b) Golden Champlain, susceptible, 60 to 60 day melon.
- (c) Extra Early Hackensack, susceptible, 82 day melon.
- (d) Iroquois, resistant 90 day melon.
- (e) Delicious 51, resistant 70 day melon.
- (f) Delicious, susceptible 70 day melen.
- (g) Improved Hearts of Gold, susceptible 75 day melon.
- (h) Early Knight, susceptible, 80 day melon.
- (i) Improved Rocky Ford Jr., susceptible 80 day melon.
- (j) Extra Early Hanover, susceptible 70 day melon.
- (k) Oka, susceptible 67 day hybrid melon.
- (1) Hearts of Gold, susceptible 85 day melon.
- (m) 3 1. a hybrid variety obtained from Vineland Horticultural Station.
- (n) 38 3 51, a hybrid variety obtained from Vineland Horticultural Station. Resistant parents.
- (o) 9 201, a resistant hybrid variety obtained from the Science Service Laboratory, Harrow, Ontario.

#### (2) Watermelon

- (a) Dixie Queen, resistant variety.
- (b) Peerless, susceptible variety.

The technique employed for planting the melons was similar to that of the melon growers in the Aldershot area. Pint size berry boxes of sterilized greenhouse compost soil were planted with 12 seeds per box. Twenty-four days later these were thinned to 6 per box and a week later the melons were transplanted to a naturally infested field. The history of the muskmelon wilt on this field was established as being at least 6 years in

duration, and repeated isolations of the organism were made by the writer over a two year period. The soil type was a fine sandy loam.

Two rows of fifteen hills were planted for each variety tested. The rows were placed in a manner calculated to expose each variety to two different areas of the field. Four plants were left per hill at final thinning.

During the seven week period immediately after planting in the field, the plants were constantly checked for signs of wilt. The early summer was unusually cool and damp, causing slow maturity of the plants.

The results recorded in Table XVIII point out several interesting things. The most important is that Iroquois is no longer a resistant melon. This was reported previously by Eide and Makila (3) and was first noted by this writer 2 years ago. Delicious 51 bred at Cornell did not appear resistant, nor did variety 9 - 201. The Oka melon showed a remarkable degree of resistance considering it is not listed as a resistant type. Extra Early Hackensack gave similar results to that of Oka. It was also noted that both watermelon varieties were not attacked.

TABLE XVIII: Comparison of Resistance of Certain Melon Varieties
to <u>Fusarium</u> Wilt of Muskmelons Under Field Condition.

TYPE OF PLANT	NUMBER OF PLANTS	NUMBER OF PLANTS	CROP
	SURVIVING	DEAD AFTER 7 WEEKS	AIELD
Benders Surprise	100	68	Poor
Golden Champlain	102	76	Poor
Extra Early Hackensack	98	33	Good
Iroquois	90	46	Moderate
Delicious 51	90	48	Moderate
Delicious	86	72	Poor
Improved Hearts of Gold	89	53	Moderate
Early Knight	96	40	Moderate
Improved Rocky Ford, Jr.	94	54	Moderate
Extra Early Hanover	95	33	Good
Oka	93	19	Good
Hearts of Gold	96	80	Moderate
3-1	92	74	Moderate
38-3-51	94	24	Good
9-201	87	69	Poor
Dixie Queen	96	0	Good
Peerless	88	0	Good

#### SUMMARY

- Of 270 isolates of <u>Fusarium</u> from muskmelon compared morphologically, only 156 resembled the strain designated as standard.
- 2. Of 99 isolates of <u>Fusarium</u> from watermelon, only 18 resemble the watermelon standard strain.
- 3. The isolates caused from 0 to 100% mortality on three hosts, and the degree of pathogenicity for isolates placed in the same morphological group was of the same order.
- 4. Certain isolates of <u>Fusarium</u> from muskmelon were found to attack watermelon seedlings to a varying degree, and vice versa.
- 5. Heterothallism did not appear to exist among the isolates tested.
- 6. Ultra-violet light, Caffeine, and Theophylline appeared to cause mutations in <u>Fusarium</u> of somewhat the same types.
- 7. The isolates were found to vary physiologically from one and other in their ability to assimilate different carbon and nitrogen sources.

  Sporulation varied with the source tested.
- 8. Certain melon varieties previously regarded as resistant, have been found susceptible in the Aldershot area.

#### DISCUSSION

The results of the present studies indicate that variation in nature is a normal occurrence in the fusariwa wilts of watermelon and muskmelon. The variants range from highly pathogenic to non-pathogenic forms, and can be fitted into groups on the basis of their morphological characters in culture. This variation in pathogenicity is in agreement with results reported by Sleeth (20), who found a wide range of variation among isolates of the watermelon wilt Fusarium from various areas of the United States, which he considered to occur naturally. This was shown by work of Hendrix et al (7). and the work of Armstrong et al (1). Armstrong et al found marked differences in pathogenicity and cultural characteristics between isolates of Fusarium vasinfectum Atk. from different localities. Snyder (22) in his work found similar results with Fusarium orthogeras App. and Wr. Miller raises strong objections to these results. pointing to the fact that many of the authors reporting this phenomenon. did not purify their cultures by single spore isolation, immediately after isolating the organism, or they did not maintain their stock cultures in a manner which would prevent mutations. McKeen (13) supports the work of Miller with respect to the muskmelon organism, but reports isolation of three natural variants of the watermolon organism which differ strikingly in pathogenicity. During this work the writer isolated and maintained cultures in the manner described by Miller (16) and followed by McKeen. It is thus surprising that the results indicate the presence of naturally occuring variants, which differ from each other with respect to pathogenicity towards three host plants and morphology. It is further interesting that isolations

were made from areas tested by Miller and McKeen. The probable reasons for Miller failing to find morphological variants are discussed previously. The fact that the writer found such variation in pathogenicity might be explained on the basis of the greater number of isolates tested.

The results of this work increases the number of <u>Fusarium</u> species known to have more than one wild type in nature. It is felt that this should not be overlooked in considering its taxonomy.

Leach and Currence (11) found the muskmelon and watermelon pathogens could be distinguished on the basis of host relationships, and this could probably be done with respect to all natural variants of each of the two pathogens. The results of the carbon and nitrogen assimilation tests. would seem to indicate that other physiological tests could be utilized in taxonomy of Fusarium. Physiological tests are of prime importance in bacterial taxonomy. Yeast classification is becoming more dependent on physiological characters, and recent work by Wickerham utilizes 37 carbon sources in this regard. With respect to the results of the assimilation tests the writer feels there is definite merit in investigating the possibility of setting up a taxonomic system in Fusarium using morphology and physiology. Classification could still be based on macrospores, which are of prime importance in the Wollenweber and Reinking (25) classification. The isolates, however, would be grown on carbon sources which stimulate the formation of macrospores. This would eliminate repeated subculturing, and taxanomic confusion due to mutations in culture.

Gordon (5) obtained the perfect stage of certain cereal rots caused by species of <u>Fusarium</u>, but the pairing experiments did not demonstrate heterothallism amongst the isolates tested. In spite of the mutagenic

agents used the perfect stage was never detected. On several occasions unique structures were obtained which might have been perithecial primordia, but this could not be definitely established. However, it is interesting to note that the type of mutations resulting from the various agents were similar.

During visits to melon growers it was noted that in the Aldershot area Iroquois was becoming susceptible. On one occasion Hearts of Gold was seen growing beside Iroquois, and the Iroquois was the most severely diseased. On many occasions healthy watermelons were observed growing beside diseased muskmelons and vice versa. These observations were substantiated in the field resistance experiment. Hearts of Gold was not more susceptible than Iroquois, but Iroquois was definitely seriously diseased, more than 50% of the plants being seriously affected. This verifies the fact reported by Eide and Makila (3) that Iroquois, a wilt resistant muskmelon variety developed by Munger (18) has become susceptible. Delicious 51, developed more recently by Munger (19) cannot be regarded as resistant on the basis of the writer's field experiment. This raises the question of whether plant breeders, breeding for resistance to muskmelon and probably watermelon wilts, are breeding against a limited number of races of the pathogens.

The writer recommends that in breeding for resistance to <u>Fusarium</u> wilt of muskmelons, new lines of melon should be tested in as many infested growing areas as possible. This would ensure the exposure of new varieties to a maximum number of pathogenic races of the organism before they are released. This probably applies to watermelons as well.

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Plates 1 and 2 show the type of spores produced by isolate 130, on 8 different carbon source media.

## Plate 1

Figure 1 Glucose-1-phosphate

Figure 2 Glycerol

Figure 3 D-Mannose

Figure 4 Saccharose

Plate II

Figure 1 Glycine

Figure 2 Histidine

Figure 3 d 1 Alanine

Figure 4 Fructose-1, 6-diphosphate

Magnification 1300 times

FIG.4

FIG.I

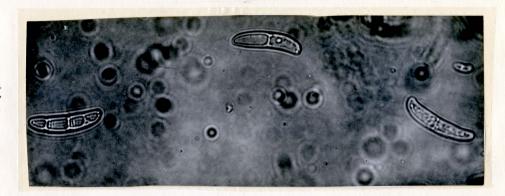


FIG.2



FIG.3

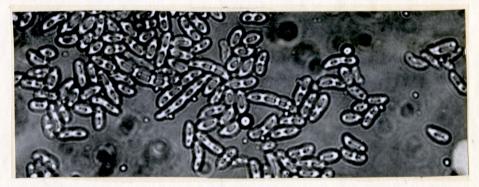


FIG.4



FIG. I



F16.2

Fig. 162111estrate appearance in culture of strains 263, respectively of the muskwelon wilt Fuseries being compared with strain I. Strain I is vertical.



FIG. I

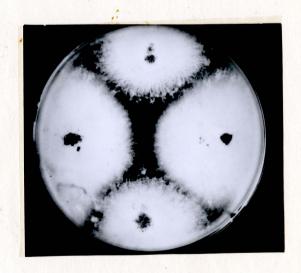
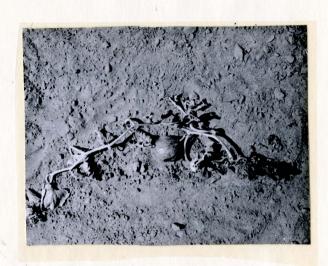


FIG.2

Fig. I & 2 illustrate appearance in culture of strains 3 & 5 respectively, of the watermelon wilt Fusarium being compared with strain I. Strain I is vertical.



FIG. X



F16.2

Fig. I shows cracking of the stem, of a Bender's Surprise melon a susceptible variety. This is a typical symptom of the disease.

Fig. 2 shows a suskeplon plant, variety Iroqueis showing severe symptoms of fusarium wilt.

FIG.I



F16.2

Fig. I G 2 show a muskmelon plant variety Iroquois, showing gummy exudate from the stem, a symptom of the disease. Note the development the melon. The fruit is almost mature, but will be unmarketable.

The map opposite, indicates the areas in southwestern Ontario, from which isolate collections were made.

