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CUCUMBER SCAB CAUSED
BY
CLADOSPORIUM CUCUMERINUM

THESIS FOR DEGREE OF M. S.

SEARS P. DOOLITTLE

1915

THESIS

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Thesis for the Degree of Master of Science.

Michigan Agricultural College.

Sears P. Doolittle

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1. The first part of the document discusses the importance of maintaining accurate records of all transactions and activities. It emphasizes that proper record-keeping is essential for transparency and accountability, particularly in financial reporting and compliance with regulatory requirements. This section also highlights the role of internal controls in preventing fraud and ensuring the integrity of the data.

2. The second part of the document focuses on the implementation of robust risk management strategies. It outlines various risk assessment techniques and provides guidance on how to identify, measure, and mitigate potential risks. The text stresses the need for a proactive approach to risk management, where risks are identified and addressed before they become significant issues.

3. The third part of the document addresses the importance of effective communication and collaboration within the organization. It discusses the benefits of clear communication channels and the role of leadership in fostering a culture of transparency and teamwork. This section also touches upon the importance of stakeholder engagement and the need to maintain open lines of communication with external parties.

4. The fourth part of the document discusses the role of technology in modern business operations. It explores how digital tools and automation can streamline processes, improve efficiency, and enhance data security. The text also addresses the challenges associated with technology adoption, such as data privacy concerns and the need for employee training.

5. The fifth part of the document provides a summary of the key findings and recommendations. It reiterates the importance of the discussed topics and offers practical advice for implementing the suggested strategies. The document concludes by emphasizing the need for continuous improvement and regular monitoring of the organization's performance and risk profile.

CUCUMBER SCAB CAUSED BY CLADOSPORIUM CUCUMERINUM.

IMPORTANCE OF THE DISEASE.

The scab or "black spot" of cucumbers caused by *Cladosporium cucumerinum*, Ell. & Art., was first noted in the United States about 1887 and since that time has been reported as generally prevalent in both this country and Europe. It has appeared at various times in nearly all parts of the United States where large quantities of cucumbers are grown and is also quite common in England and Germany.

The disease is found both in the field and in the green house but is more especially connected with the growing of cucumbers for pickling, since the young fruits used for this purpose are most susceptible.

During the past two seasons it has been very severe in the States of Michigan, Indiana and Wisconsin. This territory is the center of the pickle growing industry and the crop reaches a high valuation each year.

While other diseases have proven serious, it was estimated last year that the largest company operating in this district lost far more by this disease than any other insect or fungus trouble. The disease is to some extent a new one and no effort was made by the growers to combat it, hence it has spread rapidly and much of the crop last season was destroyed.

The nature of the fungus causes it to spread from field to field with great rapidity and the young fruits being the part most subject to attack makes the loss severe.

This is not all due to the actual fungus injury however, but also to the fact that the scab lesions give a point of entrance to various soft rots. When the cucumbers are mixed together after picking these rots will affect the sound fruits as well and they are useless for pickling purposes.

The difficulty and expense of sorting out all damaged fruit, a thing which the average grower will not do thoroughly, has led many salting stations to refuse all the fruit from fields which seriously infested with the fungus. This greatly increases the loss to the grower since he has no other means of disposal of the crop.

The disease was particularly severe last season at Big Rapids Michigan, and in the northern part of the State in general. In this locality the yield was greatly reduced although there are no figures available. The growers are beginning to realize that it is uncertain whether they can much longer grow pickles at a profit and are slow to contract for more acreage. This falling off is watched with anxiety by the companies having stations in the district since if it continues they will be obliged to change their locations and expense of such a step would be a large loss, both to them and the growers since a large item in the farm profits would be removed.

HISTORY.

The disease was first found in this country at the Geneva Experiment Station, in 1887,^{*} when it was noted by J.C.Arthur, who described it as probably being a Cladosporium.

It was first studied by him also, at the Indiana Experiment Station in cooperation with J.B.Ellis of New Jersey, in 1889[#]. The disease was very severe in that year and investigation proved it to be due to a new species of Cladosporium, which they named Cladosporium cucumerinum.

Arthur describes it as appearing first as a sunken spot with a velvety surface, from which there may be a gummy exudation, giving the appearance of an insect injury. As the disease progresses a small cavity is formed beneath the fungus covered surface. He states that this is due to the action of the mycelium which dissolves the cell walls, thus causing the exudate of gum on which the fungus subsists.

No other work was done except the description of the main symptoms and characters, control measures not being discussed.

* 6th. N.Y. Agr. Exp. Sta. Report. 1888.p.316.

Bull. 19. Indiana Agr. Exp. Sta. 1889.p.8

Frank found the disease in a garden near Berlin in 1892[#] and his description is identical with that of the fungus found by Arthur. Not having heard of Arthur's work he called it *Cladosporium cucumeris*.

He stated that it was quite prevalent in Germany at that time and noted that the growers were using Bordeaux mixture to combat it, with very little result. This fact led him to experiment with the effect of Bordeaux mixture on the spores of the fungus.

A portion of the infected material was taken and immersed in a 2 % Bordeaux for two hours, taking care that the spores were actually in contact with the fungicide. After this period they were washed thoroughly and their power of germination tested. He found that the copper apparently was not fungicidal in its action toward the spores as they germinated readily. However, no work was done beyond this one experiment.

Humphrey noted the disease in Massachusetts as affecting the leaves in the season of 1892[‡]; no other part of the plant being attacked. The leaves are described as showing translucent, watery spots, the fungus growing within the tissue and sending out hyphal branches through the stomata, on which the conidia were produced.

Frank A.B. Krank. d. Pflanzen. 1892, p. 30.

‡ Humphrey J.E. 10 th. Rept. Mass. Exp. Sta. 1892 p. 222.

He describes the spore production as follows:-

" A highly complicated mass of spore threads, consisting of the successive acropetal production of sprout buds from the originally terminal joints so that there are formed chains of successively smaller and smaller cells".

He simply described the organism without recommending any control beyond the suggestion of spraying.

In 1896[#] Aderhold noted the fungus near Breslau in Germany and identified it as *Cladosporium cucumerinum*. He demonstrated its parasitism by inoculation experiments and found it prevalent both in the field and under glass.

He endeavored to connect it with *Sporidesmium mucosum*, found on other cucurbits but did not find sufficient ground to warrant such a belief.

Since that time the literature shows little work on the disease, being confined to mere mention of its occurrence. There is no record of special experiments for its control except the work of Frank mentioned above. I have it verbally however, that the Department of Agriculture conducted spraying experiments along this line in Michigan about 1905 with little success, the work not being published.

[#] Aderhold R. Krank. d. Pflanz. VI. 1896. p. 72.

SYMPTOMS ON THE HOST.

On the fruit the disease appears as a very slightly sunken, water-soaked spot, usually accompanied by a small drop of gummy exudation. As the fungus progresses this spot becomes more sunken and is covered with a smooth, velvety, olive black coating of spores and conidiophores. These spots are seldom more than 2 - 3 mm. in diameter, although several will often coalesce to form larger patches.

If cut through in cross section the fruit shows a shallow depression of about .5 - 1 mm. in depth. This depression is underlaid by a slight cavity in the flesh but the lesion never extends deeply into the fruit. Where the fruit is allowed to mature however, these spots may cause a cracking of the flesh or the cucumbers may be gnarled and deformed.

On the young fruit the infection seems to occur principally at the blossom end, there often being a number of spots at this point. This fact may be due to the blossom holding drops of moisture at this end of the fruit and thus allowing more chance of infection.

In the field the fungus will produce typical lesions in from two to three days under favorable weather conditions, i. e. heat and moisture. It was found by experiment that many of the spots which were unnoticeable at the time of picking would develop after the fruit had been in the crate



for 24 hours, since the mass of pickles generates a certain amount of heat and moisture, thus furthering the growth of any fungus present. This fact explains some of the difficulty experienced in trying to cull out all infected fruits from diseased fields.

On the stem the lesions appear first as a slight crack in the surface, accompanied by a gradual splitting if growth is rapid. These lesions often attain a length of from .5 - 1 cm. but are usually only from 4 - 6 mm. long. They occasionally extend rather deeply into the stem and may cause it to break off but ordinarily the injury is superficial. The surface of the spot is not covered with spores as it is in the fruit but has more of a brownish, roughened appearance. Spores are present together with the conidiophores but do not produce a dense felt as on the fruit.

The character of the disease on the petioles is exactly similar to that on the stem. In both cases the lesions are slower to develop than on the fruit, a period of 5 to 7 days being required before typical lesions occur under ordinary conditions.

On the leaves the fungus is not as common as on the parts of the plants discussed before. Where it appears we find it as a brown, translucent spot which tears out very easily. These spots may develop soft rots and become merely a watery mass if in contact with the soil. The lesions may take in as much as half the leaf or be only small patches which often tear out in the case of young and rapidly growing leaves.

The writer noted only one typical case of leaf infection last season and that was under conditions particularly favorable for fungus growth. The disease does not seem to appear until the vines are well matured, no record of any early infection being found last season. In the Big Rapids district where the disease was most severe, it did not commence to any extent until the first week of August and this seems to have been the case throughout the State.

DESCRIPTION OF THE FUNGUS.

The causal organism, *Cladosporium cucumerinum* was isolated from diseased fruits sent to the writer from Big Rapids. Reinnoculations into healthy cucumbers produced typical lesions in about five days, and from these the fungus was reisolated, thus proving its pathogenicity.

The organism is a Hyphomycete and belongs to the small group of *Cladosporiums* which are truly parasitic in nature.

It consists of a much branched, closely septate granular mycelium, which is occasionally enlarged and swollen so that the segments appear nearly globular. This last character is much more common on certain culture media than on the plant.

The conidia are borne at the apex of the fruiting hyphae, being formed by a simple cutting off process. The hyphae are more slender than the vegetative mycelium and not so much septate. The conidia occur in chains and are occasionally two celled though this not common, most of the spores being single celled. They vary in size from 3 - 4 microns in diameter and 7.5 in length to 3 - 4 by 12 microns. They are ordinarily rather a broad oval, sharply pointed at one end but in the case of the two celled forms we find them more cylindrical in shape. The chains are very easily broken and the spores detached so that it requires careful handling to observe them undisturbed.

On germinating the spore sends out a rather large germ tube which may come from either end of the conidium or from both. This becomes septate soon after its appearance, being many septate under normal conditions. The branching is very profuse, the branches originating directly back of the septa and in some cases occurring from nearly every cell. The mycelial strands occasionally show a fusion but this uncommon and the writer has observed it in only two cases.

Spore production takes place very soon, usually in two days on ordinary media. The fruiting hyphae begin to cut off into spores, at the apex of each tip if the hyphae be branched, and the spores continue to bud off into more conidia. There soon is produced a very complicated chain of conidia, the whole cluster forming a complex branched mass.

As said before these are very easily broken and in some of the older works we see the fruiting hyphae shown as having only the remnants of the spore chains still adhering to the conidiophore.

REACTION OF THE ORGANISM TO VARIOUS CULTURE MEDIA.

The fungus was grown on 14 separate media, the formulae for which are given at the end of this paper. Eight tubes of each sort were used, these being divided into two lots of four tubes each. Two of each four were sealed hermetically and the others left with ordinary cotton plugs. One set was kept in perfect darkness and the other allowed to remain in the light, both being at room temperature.

The two sets were examined at intervals of 2, 5, 7, 9, 15 and 24 days respectively, the following characters being noted:-

Spore production.	Color of growth.
Discoloration of medium.	Character of spores.
Rate of growth.	Character of mycelium.

All colors are from comparison with the Ridgway Color Charts. The mycelium and spores were examined only in the unsealed tubes as it was impossible to gain access to the those sealed.

CUCUMBER AGAR.

Light.Unsealed tubes.

Spore production after 2 days.

Medium not discolored.

Growth rapid.

Colon Light yellow olive changing to dark olive.

Spores mostly 1 celled, 3 x 7 microns.

Mycelium much branched, many septate. Conidiophores short.

Sealed tubes.

Appearance the same as unsealed at first.

Medium not discolored.

Growth slow, ceasing after 9th. day.

Final color of colony, buff olive.

Dark.Unsealed tubes.

Spore production in two days.

Medium not discolored.

Growth slightly more rapid than in the light.

Color same as that in the light.

Spores and mycelium same as in the light.

Sealed tubes.

Color of growth buff olive.

Medium not discolored.

Growth slower after 2nd. day, ceasing by the 9th. day.

1 1/2 2

NUTRIENT AGAR.

Light.Unsealed tubes.

Spore production after 2 days.

Medium not discolored.

Growth mediumly rapid.

Color light olive green with light buff border.

Spores mostly one celled, 4 x 7.5 microns.

Mycelium much branched and segments often swollen.

Sealed tubes.

Color and appearance of colony same as above.

Medium not discolored.

Growth slow, ceasing about 9th. day.

Dark.Unsealed tubes.

Spores produced in 2 days.

Medium not discolored.

Growth slower than in light.

Color dark olive.

Spores same as in the light.

Mycelial growth same as in the light.

Sealed tubes.

Appearance and color same as in unsealed tubes.

Growth slow after 2nd. day, ceasing by the 15 th.

Medium not discolored.



HARD POTATO AGAR.

Light.Unsealed tubes.

Spores produced in small numbers after 2 days.

Medium not discolored.

Growth slow.

Color olive green.

Spores 3 x 7 microns, one celled.

Mycelium slender, not much branched, conidiophores short.

Sealed tubes.

Color and appearance of colony same as in unsealed.

Medium not discolored.

Growth slow, ceasing after 9th. day.

Dark.Unsealed tubes.

Spores produced after 2 days.

Medium not discolored, until last observation, when it was dark.

Growth slow, but slightly more rapid at first than in light.

Color dark olive green.

Spores and mycelium same as in the light.

Sealed tubes.

Appearance and color same as unsealed.

Growth ceased after the 7th. day.

Medium not discolored.

CORN MEAL AGAR.

Light.Unsealed tubes.

Spores produced in large numbers by the second day.

Medium not discolored.

Growth fairly rapid.

Color. Slate olive.

Spores 3.5 x 7 - 8 microns.

Mycelium not closely septate.

Sealed tubes.

Color and appearance of colony same as in the unsealed tubes.

Medium not discolored.

Growth ceased after 9th. day.

Dark.Unsealed tubes.

Spores same as in the light, produced after 5 days.

Medium not discolored.

Growth same as in the light.

Color. Light gray olive.

Spores and mycelium appear same as in light.

Sealed tubes.

Same as unsealed tube in all respects.

OAT AGAR.

Light.Unsealed tubes.

Spores numerous after 2 days.

Medium not discolored.

Growth rapid.

Color. Dark slate olive with lighter border.

Spores 2.5 - 3 x 7 microns. Mostly one celled.

Mycelium not very luxuriantly branched, many septate.

Sealed tubes.

Color and appearance the same as above.

Medium not discolored.

Growth ceased in 15 days..

Dark.Unsealed tubes.

Spores numerous.

Medium unchanged.

Growth rapid.

Color. Dark slate.

Mycelium same as in the light, conidiophores more sparse on the surface.

Sealed tubes.

Identical with unsealed in appearance but all growth ceased in 5 days.

CUCUMBER STEMS.

Light.Unsealed tubes.

Colony whitish gray in color.

No spores, until 5 th. day.

Medium blackened

Spores 3 x 7 microns.

Mycelium very much like that on cucumber agar. Conidiophores are borne on very long hyphae.

Sealed tubes.

Same as the unsealed in all appearance.

Growth ceased after the 5th. day.

Medium blackened.

Dark.Unsealed tubes.

Spores numerous after 5 days.

Medium unchanged, till last observation when it appeared dark.

Growth very slow.

Color. Dark gray.

Mycelium same as that in the light.

Spores same as those grown in the light.

Sealed tubes.

Very slow growth.

Conidiophores very short and sparse.

Color same as in unsealed tube.

Medium blackened and shrivelled in appearance.



CARROT PLUGS.

Light.Unsealed tubes.

Spores very numerous after 2nd. day.

Medium much discolored.

Growth very rapid.

Color. Dark gray green changing to dark green olive.

Spores often 2 celled, 3.5 x 7 microns to 4 x 12 microns.

Mycelium slender not so much septate as in some cases.

Sealed tubes.

Spores seem to be produced after 2nd. day.

Medium not as much discolored as in unsealed tubes.

Growth slower than in unsealed, stopping about 15 th. day.

Color. Light olive green with lighter border.

Dark.Unsealed tubes.

Spores produced in large numbers by 2nd. day.

Medium darkened as in light but not as rapidly.

Growth rapid, being faster than in light.

Color. Deep olive green.

Spores and mycelium same as in light.

Sealed tubes.

Very slow growth.

Medium not much discolored.

Color. Buff olive with grayish border.

POTATO PLUGS.

Light.Unsealed tubes.

Spores not produced in large numbers till the 5th, day.

Medium darkened slightly.

Growth slow.

Color. Deep olive with light border.

Spores 4 x 6 microns, mostly one celled.

Mycelium similar to that on the carrot.

Sealed tubes.

Medium darkened.

Growth slow.

Color. Deep olive.

Dark.Unsealed tubes.

Spores produced in large numbers.

Medium slightly darkened.

Growth rapid.

Color. Olive green.

Spores produced of same type as on carrot. Mycelium also s similar to that on carrot.

Sealed tubes.

Medium much darkened.

Growth very slow after the 5th. day.

Color. Buff olive with lighter border.

CELERY STEMS.**Light.****Unsealed tubes.**

Spores produced in 2 days.

Medium discolored slightly.

Growth rapid.

Color. Buff olive.

Spores 3.5 x 8 microns., one celled. Conidiophores short.

Mycelium of large diameter many septate, segments swollen.

Sealed tubes.

Very slight discoloration of the media.

Growth very slow, ceased after 15 days.

Color. Dark olive buff with gray border.

Dark.**Unsealed tubes.**

Spores produced in large numbers.

Medium blackened.

Growth very rapid.

Color. Light olive green.

Spores and mycelium of same type as in the light.

Sealed tubes.

Medium not much darkened.

Color. Same as unsealed, in the dark.

Growth slow.

RICE..**Light.****Unsealed tubes.**

Spores not produced until the 5th. day.

Medium has reddish tint.

Grows fairly rapidly.

Color. Dark slate gray.

Spores 3 x 7 microns.

Mycelium much swollen in older segments, more than on any other medium used, some of the cells being almost spherical.

Sealed tubes.

Medium not discolored.

Growth very slow.

Color same as in unsealed tube.

Dark.**Unsealed tubes.**

No spores formed until 9th. day.

Medium slightly reddish in color.

Growth very slow.

Color. Dark gray.

Mycelium same as that above. Spores are all one celled and similar to those in the unsealed tube.

Sealed tubes.

Medium slightly darkened.

Growth very slow.

Color. Grayish black.

CORN MEAL.

Light.Unsealed tubes.

Spores produced in large numbers.

Medium not discolored.

Growth rapid.

Color. Deep grayish olive.

Spores same type as on rice.

Mycelium is many septate and much branched.

Sealed tubes.

Medium not discolored.

Growth very slow.

Color. Light olive green.

Dark.Unsealed tubes.

Spores produced in large numbers on the second day.

Medium not discolored.

Growth rapid.

Color. Dark olive green.

Spores and mycelium same as those described above.

Sealed tubes.

Medium unchanged in color.

Growth much slower than in unsealed.

Color the same.



PARSNIP.**Light.****Unsealed tubes.**

Spores produced in large numbers.

Medium blackened.

Growth very rapid.

Color. Olive green.

Spores and mycelium identical with those on the carrot.

Sealed tubes.

Growth slower than in unsealed tube.

Color. Light olive.

Dark.**Unsealed tubes.**

Spores produced on the 5 th. day.

Medium not discolored.

Growth slower than in light.

Color. Light olive green.

Spores and mycelium the same as those in the light.

Sealed tubes.

Medium is not discolored.

Growth much slower than in unsealed tubes

Color. Light buff olive.

From the examination of the above data it can be seen that the fungus behaves very much the same on all media. The most startling fact in the experiment is the rapidity with which heavy spore production occurs. The usual time was two days and in many cases a large growth had taken place in that interval.

The best medium seems to be either carrot or parsnip plugs followed by cucumber agar. These all furnish excellent media for the development of the organism with rapidity.

The slight variations in color are numerous but never such that the typical olive shade is not present. The rate of growth was very different in the different media and the fact that the fungus does not seem to show ability to grow under anaerobic conditions, since all the culture in the sealed tubes had their growth very much checked. The spore form does not vary appreciably nor is the character of the mycelium much altered on any of the media used. There is evidently little danger that the fungus cannot be easily identified on any media used.

GERMINATION STUDIES.

Van Tieghem cells were employed for this experiment, using hanging blocks of the various media, taking those only which had a very slight inoculation of the fungus spores. These cells were kept at room temperature and germination was followed closely, camera lucida drawings being made of any important features of the growth.

Nutrient Agar.

Germination on this media took place in 20 hours. Growth was slow until 30 hours old when the mycelium began to branch profusely and soon filled the entire drop of media with a closely interwoven mass of fungus threads.^o The older mycelium showed the cells formed by the septa as much swollen some of them being almost spherical.[#]

On the eighth day after starting the experiment it was found that in a few isolated instances the hyphae had cut off to form chains of conidia. These increased in length and became very complex in their manner of branching. Nearly all of the conidia were one celled, very few of the two celled form being found.

Cucumber Agar.

Germination in this case took place after 26 hours. At the end of 36 hours the growth became rapid and the mycelium became much branched, being many septate, with lateral branches after almost every septum in some cases.

[#] Plate I. fig. 5. ^o Plate II. fig. 1 - 5.



After about three days the mycelium shows a swollen appearance in the older portions, but this is not so pronounced as on Nutrient agar.

Conidia were produced in large numbers and were identical in appearance with those on the other media.

Hard Potato Agar.

Germination on this media took place in 28 hours. Growth was very slow at all times, mycelium being closely septate but not much branched and the growth scanty.

Spores were produced in small numbers at about 3 days old. Some of the germ tubes cut off into conidia when only 40 hours old and before any branching had occurred. All conidia were identical with those on the other agars.

Corn Meal Agar.

Germination took place after 32 hours. Mycelium grew quite rapidly after about 40 hours but was as not as luxuriant as that on cucumber agar. Mycelium slender and and not as closely septate as the other growth on the various media.

Conidia were produced in short, only slightly branched chains after six days. The spore masses were much less in complex in this case than on the other agars.

Distilled Water.

Spores germinated after 30 hours but grew slowly for 48 hours after which growth ceased. No conidia were noted.



RESISTANCE OF THE FUNGUS TO DESSICATION.

A number of sterile cover glasses were used in this experiment, each one being smeared with a solution of spores of *Cladosporium cucumerinum* in distilled water. These were allowed to dry in the light and at room temperature and were then broken up with sterile forceps and the fragments planted in tubes of cucumber agar at intervals of 24 hours.

Five tubes were inoculated in this way each day and the experiment continued till there was no chance of further germination taking place.

The cultures germinated up to the 18th. day but after that time no colonies developed. A duplicate of the experiment was run later and in this case the spores retained their vitality for 19 days.

While the resistance to dessication on glass seems to be only from 18 to 19 days, yet the conditions in nature are probably much different. The various oils, gums, etc. found on the seed coats, fruit and plant itself will often protect the spores lodged thereon from the effects of dessication for long periods. This has been proven true with other organisms and may very likely be the case with *Cladosporium cucumerinum*. Therefore the above data cannot be taken as fixing the length of resistance to drying absolutely.

THERMAL DEATH POINT.

The apparatus for this experiment consisted of an iron vessel filled water and agitated by a small motor-driven paddle. A thermostat regulator attached to the gas burner beneath kept the water at the desired temperature and the tubes were suspended in it in a wire basket.

The temperature was taken from a thermometer placed in a test tube of the same size and thickness and containing the same amount of media as those in which the spores were placed. These were 18 mm. in diameter and .5 mm. in thickness.

The temperature of the bath was held at the point desired from 3 to 5 minutes before placing the culture in the water. When the temperature seemed correct the tube of melted agar was placed in the bath for 3 minutes to bring it to the required temperature and was then removed, quickly inoculated and placed again in the bath for exactly 10 minutes, when it was taken out and plated at once.

Three sets of tests were run going from 36 C. to 58 C. The thermal death point in all these seemed to be at 50 C. and will be considered here as such.

This temperature will allow the fungus to thrive in any climate where the cucumbers are at present grown for market purposes and removes any possibility of hot weather greatly reducing the vitality of the organism in this climate.

MAXIMUM, MINIMUM AND OPTIMUM TEMPERATURES.

The apparatus for this experiment consisted of a metal box having a compartment in one end filled with ice and one at the other end with water kept heated by an incandescent lamp. The space between the two was divided into compartments, thus getting varying degrees of temperature as follows:-

8° C.
11° C.
19° C.
24° C.
29° C.
34° C.
43° C.
51° C.

Test tubes of cucumber agar inoculated with the fungus were placed in each of these compartments and the amount of growth if any observed.

Tube.	Temperature.	Amount and Character of Growth.
1	8° C.	None.
2.	11° C.	Very slight growth.
3.	19° C.	Fairly heavy growth.
4.	24° C.	Heavy growth.
5.	29° C.	Very slight growth.
6.	34° C.	Bare trace of mycelium.
7.	43° C.	None.
8.	51° C.	None.

From this data the maximum temperature seems to be about 35° C., the optimum about 25° C. and the minimum about 10° C.



EFFECT OF WINTERING ON THE SPORES.

Trash collected from a badly infected field near Big Rapids was kept outside during the entire winter up to April when it was brought into the laboratory and examined for traces of the fungus.

The lesions on the old cucumber remains were closely examined but no signs of spores appeared under the microscope. The same proved true of the stem lesions but the wintering process had been so severe that it was almost impossible to recognize many of them.

Some of the stems and remains of cucumbers were washed thoroughly in a flask and the surfaces scraped with a scalpel. This resulting suspension was then centrifuged and the precipitate examined carefully under the microscope. No results were obtained from this and some of the material was then plated out on agar but nothing similar to *Cladosporium* developed.

Another method was then tried since the other yielded no results. Stems and fragments of cucumbers were ground in a large mortar with quartz sand and water, pulverizing the material thoroughly. The resulting suspension was taken and dilutions plated from it as follows:-

1 - .100 1 - 10,000 1 - 1,000,000

This work gave no results however as the mass of bacteria which developed prevented observation.

Later an attempt was made to grind up merely the actual remnants of lesions and plate from those, using higher dilutions, hoping thus to avoid such heavy contamination.

The dilutions were made as follows :-

1 - 1000 1 - 100,000 1 - 1,000,000, thus getting very high dilutions. Since the chance seemed small of any spores of the fungus being found in only one set of plates, six were made, which, while the bacteria were not so evident as before, gave no sign of the Cladosporium

These results do not prove that the fungus does not winter over however, as the writer had little material for examination and on that account cannot speak definitely in this regard.

It seems likely however, considering the nature of the fungus, that it does winter over in the last year's rubbish. Which is usually left where it lays during the winter.

RELATION OF THE ORGANISM TO TEMPERATURE AND HUMIDITY.

These data were taken from the field notes of H.J.Buell who conducted spraying experiments for the H.J.Heinz Co. at Big Rapids in 1914.

The date of infection comes from his data and while not absolutely correct it is accurate enough for the purpose. The meteorological data are from the United States Weather Bureau Reports.

Plot.	Infection.	Date of Last Rain	Amt. of Rain.	Av. Temp.
		Previous.		Following.
1.	Aug. 29.	Aug. 18 - 20.	1.78	82°
2.	" 7.	" 1	.55	81°
3.	" 17	" 9 - 10	.36	78°
4.	" 17	" 9 - 10	.36	78°
5.	" 26	" 18 - 20	1.78	82°
6.	" 16	" 9 - 10	.36	78°
7.	" 29	" 18 - 20	1.78	82°
8.	" 19	" 16	.05	83°
9.	" 22	" 18 - 20	1.78	82°
10.	" 25	" 18 - 20,	1.78	82°

The temperatures are for the three days immediately following each rain as that is the critical period.

It will be seen that in five cases out of ten the infection ^{of the season} took place three to five days after the heavy rains of August 18 and 20, which were followed by hot weather. Three out of

the five remaining plots were infected after the rain of August 9 - 10, showing that the infection in this case was about mid August. From reports received from various localities I believe that this is true for all of Michigan, Indiana and Wisconsin.

The fungus can endure a temperature of about 35 °C. and continue to grow so that it is not checked by warm weather as is Bacterial Wilt for instance. This causes it to be peculiarly severe under mist and very hot conditions.

The fact that it grows under such conditions make its attack most severe where the crop would be largest, since the host plant is one requiring large quantities of moisture to produce large crops of pickles, the young fruits being 90 % water. This fact necessitates a rainy season to make the largest crop.

Hence where vines are on rather low lands and the humidity is high, we find the fungus damage most serious. I noted one field last season where the vines were on low land and had made so heavy a growth that the leaves completely covered the surface of the ground and the crop was a large one. One week after scab appeared in this field the writer made an examination and found 95 % of the fruit ruined, by actual count, the leaves and stems also being badly infected with the fungus. In this case the weather was unusually favorable for the fungus, but it shows how severe it may become, when conditions are favorable for attack.

AGENTS OF DISTRIBUTION.

The methods of transmission of the disease may be many. The spores are produced in great numbers on the fruit and to some extent on the stems and petioles.

They are very easily detached and can be wind blown to some distance. They are not so likely to be carried in this manner as in some other however as they are borne so close to the ground that the leaves protect them from air currents to quite an extent.

A possible means of distribution by the agency of insects, but this is a minor one compared with some others. In a field already infected the splashing of the rain would throw the spores from infected fruits to healthy ones very readily.

Probably the main cause of infection is the human agency however, since the pickers must be the carriers of great numbers of these spores which adhere to their clothes and shoes when working in the patch. These are easily shaken off in any cucumber fields through which they pass and we have a new case of infection in three or four days time.

INNOCULATION EXPERIMENTS.

These experiments were carried on in the greenhouse with plants which had reached a fair size and were not infected with any fungus so far as known.

Infectations were made in two ways as follows:-

1. Spores were sprayed on the plants with a sterile atomizer, spores being in solution in sterile distilled water. The plants were all kept under bell jars with a good supply of moisture.

2. Plants were punctured with a sterile needle after the surface of the plant had been washed with $HgCl_2$ solution. Spores were introduced into the puncture with another sterile needle and the wound covered with a tiny piece of moist cotton. All plants were kept under bell jars .

It may be said here that the writer has never been able to get infection of the plant by spraying the spores on the surface, unless there was some mechanical injury already present. This is true to some extent of the *Cladosporium* of the Peach and also of that causing the scaly bark of citrus trees and it is possible that *Cladosporium cucumerinum* can only penetrate the host by some mechanical injury.

Stem Inoculations.

Stem infection took place in practically all inoculations where punctures were made. The stem first showed a water-soaked spot at the point of inoculation after 3 or 4 days.

In 6 days to a week it became a buff brown, and a slight splitting occurred. However, the plant was not growing fast enough to cause the large split seen under field conditions. The infection did not seem to extend deep into the stem nor far from the spot of puncture and it is likely that many of the lesions seen in the field are not entirely due to the fungus but also to the tendency of the stem to crack open when injured during rapid growth.

Leaf Innoculations.

Leaf infection was secured only where the plant was actually punctured and then in only a few cases. The infected spot became lighter in color and then gradually developed a translucent, brown appearance. This spot took up about one-half the leaf and soon became shrivelled and water soaked. The spores were borne here and there on the surface but could be seen only microscopically.

The water-soaked blackened appearance so often found in the fields is possibly due to outside infection with soft rots of some sort. The tissues are softened and the cell walls more or less dissolved so that the leaf would form a very susceptible medium for such infection. The writer has never seen this condition produced in plants inoculated with *Gladosporium cucumerinum*, nor does it appear to be a constant character in the field.



Innoculation of the Fruit.

With the fruit the same factor of requiring mechanical injury held true, in the work of the writer at least. Where the spores were sprayed on the fruit the successful inoculations were relatively few while when injured they were almost universally successful.

The first sign of infection is a water - soaked spot, due to the dissolving action of the mycelium on the cells of its host. This appears about 48 hours after inoculation and becomes gradually more sunken until the fungus growth covers the surface and the entire epidermis is destroyed, being replaced by the mass of fruiting hyphae which give the olive black color to the infected spot.

The mycelium of the fungus as seen in stained sections does not penetrate the cell but exerts a dissolving action upon them. A cavity is formed beneath the epidermis and the mycelium is found pressed closely against the walls of the gradually disintegrating cells. This mycelium grows in a medium composed of the products of this cellular decomposition and in section we see the strands of mycelium massed around the edge of the cavity and intermixed with the gum and fragments of the broken cell walls. After the fungus begins to fruit on the surface and becomes mature the action of the parasite seems to be checked and the host begins to form a thin layer of cork cells about the lesion.

This seems to be resistant to the fungus action as the lesions are never more than 2 - 3 mm. deep. This corky layer is more apparent after the fruit is withered and dry, when the epidermis may be peeled off and the bare cavity will be found underlaid with a tough layer of corky cells.

When seen in cross section the fungus growth appears to be on the surface with a shallow cavity beneath, but this is not really the case as the epidermal tissue is entirely destroyed and the fungus penetrates as far as the bottom of the cavity. The formation is due to the fact that the web of mycelium hardens and dries on the surface by the evaporation of the gummy material's volatile portion and a sort of crust forms covering the cavity beneath.

The threads of the mycelium do not seem to penetrate the cells but are found closely pressed along them. The cells for three or four layers back in young lesions show traces of gradual disintegration by their broken walls, but the fungus apparently cannot actual penetrate the cell wall.

EFFECT OF BORDEAUX MIXTURE ON THE FUNGUS
IN FIELD EXPERIMENTS,

The work referred to here was done during the season of 1914 by Mr. Buell under the direction of the . . . H. J. Heinz Co. The different fields were sprayed on half and the other left as a check. The mixture used was a 3 - 6 - 50 Bordeaux, applied at the rate of 200 gallons per acre. The spraying was done in rotation, beginning with one field and spraying all the rest in regular order, then commencing again with the first.

The Big Rapids district was the center of the severe attack of scab noted last season and as none of the fields except those used for experiment, were sprayed, there were exceptional difficulties in controlling the fungus.

As some of the fields sprayed seemed to show partial control while others did not, Mr. Buell's data were examined to see if it would throw any light on the subject. It was found by comparison that there seemed to be some correlation between the time of spraying and the date of the following rain, which had an effect on the control of the disease. The Weather Bureau Reports furnished part of the data and Mr. Buell's data, the rest. The timing of the spray being purely accidental makes it more or less variable, but for the critical period it gives the relations desired.



We have both extremes, the spray applied just before a rain and just afterwards. The difference in methods is very marked in its results as will be seen below.

Plot 2.

Sprayed some time after the rains in July and 5 days before the heavy rain of August 18, when most of infection took place.

August 8.	Scab appeared.
" 15	15 % on sprayed side
	40 % " unsprayed "
August 19	25 % on sprayed side.
	40 % " unsprayed ".

The entire plot was abandoned the next week from scab.

Plot 3.

This plot was sprayed from one to three days after every rain and in no case were vines freshly covered with spray previous to heavy storms.

August 17.	Scab appeared.
" 27.	50 % on sprayed side.
	70 % on unsprayed side.

An infection of 50 % does not represent control as it is equivalent to total loss of the crop

Plot 4.

Sprayed one to two days previous to all rains in August, never more than two days previous to any rain.

August 7. Scab appeared.

" 27. 10 % on sprayed side.

70 % " unsprayed ".

September 3. No change on the sprayed side.

This plot comes very closely to representing perfect control of the fungus.

Plot 5.

Sprayed no more than three days previous to all rains in August and only a few hours before that of August 18.

Scab appeared about August 19.

August 26. 5 % on the sprayed side.

" 15 % " " unsprayed "

" 30 10 % " " sprayed side.

40 % " " unsprayed "/

This is another example of fair control of the disease.

Plot 6.

Sprayed well until just preceding mid August when the spraying was abandoned.

September 1. 40 % on the field everywhere, spray showing no effect.

Plot 7.

This field was sprayed within at least three days of all rains during the season.

Scab appeared August 15.

August 24 55 % on unsprayed side.

15 % " sprayed. "

This like the preceding shows fair control of the fungus.

Plot 8.

This was sprayed just a few hours before the wet weather of August 18 and just before all rains in August.

August 19. Scab appeared.

September 5. 15 % on the sprayed side.

75 % " " unsprayed ".

September 8. No change in percentages.

This is another example of fair control from proper timing of the spray.

Plot 11.

This was sprayed four or five days before all the important rains of mid August and the rest of the season as well.

August 24 scab appeared.

September 3. 50 % on the unsprayed and 50 % on sprayed.

From the above data it can be seen that where the spray was properly timed the results were much superior.

When the spraying was so arranged that the vines had little chance to make new growth before a period of humid weather, they were fairly well protected.

On the other hand, where the spray was applied some time before the rains, the vines were seriously affected on both sprayed and unsprayed portions.

This was in particular true in August when the epidemic commenced, as during the month of July there was so little rain that any fungus injury was impossible.

The fact that only one half of the field was sprayed and the remainder usually seriously affected added greatly to the difficulty of securing any reasonable amount of success in the work. The writer has no means of knowing how correct the percentages of infection are but as they are based on actual counts they should be fairly correct as to the amount of disease present.

When it is considered that to be effective a fungicide must so cover a plant that any spores falling on it in a drop of water will immediately come in contact with it at solution of the spray, one can see the importance of spraying as soon before a rain as possible.

The cucumber plant grows very readily and will produce young fruits continually. Hence to give full protection it would be necessary to spray at intervals of every two days, which is a manifest impossibility.

The next best thing is to make each spraying effective for the longest time possible. Hence we need not worry so much during periods of dry weather, in fact it is better not to spray at such times, since the pollination is injured to some extent at every spraying. But if the grower can study carefully the Weather Reports and local conditions, he may be able to get a fair idea of the conditions which may prevail for the next day or two and endeavor to forestall the danger periods by applying the spray as little before the rain as possible.

This theoretically the proper way to apply fungicides but it will require some effort and foresight for the grower to follow out these principles successfully.

CONTROL OF OTHER PARASITIC DISEASES CAUSED BY CLADOSPORIUM SP.

Peach Scab, caused by the fungus *Cladosporium carpophilum*[#] is found on the surface of the fruit, but does not penetrate beyond the first layer of epidermal cells to any extent, absorbing food from its host but not working into the flesh. The cells directly beneath the fungus threads usually form a cork like layer and this hardening of the surface is what causes the cracking of the fruit.

On the twigs it takes the form of small purple spots which produce conidia on their surfaces as does the fungus when on the fruit itself.

The fungus usually is controlled by self-boiled lime and sulfur but Bordeaux mixture is an effective remedy except for the fact that it is likely to injure the fruit.

- - - - -

Tomato Scab, caused by *Cladosporium fulvum*, is a disease of the leaf of the tomato, the fungus penetrating the host tissues and causing a yellowing of the upper surfaces of the leaves, the conidia being borne on the lower surface; giving it a mildewed appearance.

Bordeaux mixture is a perfect control for this disease if used early enough in the season but control when it has progressed far is difficult.^o

[#] Chester. Del. Agr. Exp. Sta. Rpt. 8, 1895 p. 60.

^o Masee. Diseases of Cult. Plants & Trees. 1913.



Scaly Bark of Citrus Fruits caused by the fungus
Cladosporium herbarum, var. *citricola*.[#]

This species affects the shoots of citrus trees and causes a flaking out of the bark in the infected portion. The fungus penetrates the cambium layer and causes the bark to become hard and brittle. A false cambium often forms beneath and the infected portion is left between two rings of wood. A gummy substance is usually formed by the destruction of the host cells.

It affects the rind of the fruit and often causes portions to drop out. The conidia are borne on the surface of both fruit and twigs as in all *Cladosporiums* noted.

Cladosporium citri or citrus scab causes a superficial injury to the surfaces of the fruit twigs and leaves of citrus fruits. It produces small wart like swellings and penetrates slightly into the tissues the spores being borne on the surface.^o

Moist weather is very essential to its growth and Bordeaux is recommended for its control as it is in the case of Scaly bark.

[#]
 Bul 106, Fla. Agr. Exp. Sta. 1911.

^o
 Bull. 8. Div. Veg. Path & Phys. 1896.

From the comparison of the above disease with *Cladosporium cucumerinum*, it will be seen that they are all similar in nature, the spores being borne on the surface of the host and the injury merely a superficial one.

The formation of a gummy product seems to indicate that they are alike in their effect on the cells of the host and we find a corky layer being formed in most cases to protect the host.

The diseases all being caused by fungi of the same type and the injuries being similar in their main characters, we must suppose that a fungicide which controls all but one of them should be at least partly effective against that one. It is hardly likely that Bordeaux mixture should be useless as a fungicide in the case of Cucumber scab, when all other *Cladosporiums* are killed by its contact with their spores. There is no reason to believe that *Cladosporium cucumerinum* is as resistant as one would think from Frank's work and the tests made with fungicides in the laboratory have shown that this resistant quality so long ascribed to it is in reality lacking.



RESISTANCE OF THE ORGANISM TO FUNGICIDES.

As no work had been done on the disease except that by Frank, it was decided to attempt to discover whether the copper sprays were as ineffective as had been supposed by most writers. Other fungicides were also tested in the hope of finding something which would prove effective.

Frank's work was also duplicated in an attempt to see if the immersion of spores in the spray really was as harmless as it seemed.

Duplication of Frank's Work.

A culture of *Cladosporium cucumerinum* on carrot was taken and a small portion of the material was immersed in a 2 % Bordeaux mixture for two hours, taking care to immerse the spores thoroughly in the spray and bring them actually in contact with the solution.

After two hours the spores were removed, washed thoroughly in distilled water after hanging in a current of gently flowing water for sometime, and then placed in hanging drops in Van Tieghem cells.

On examination of the spores after 36 hours it was found that 90 % of them had germinated indicating that Frank's work was correct. Immersion in a spray is not to be compared with conditions in the field however, since in that case the fungus has dried on the host in the presence of the various chemical agents in the atmosphere, and many

chemical changes γ have occurred which alter the value of the spray to quite an extent. This has been pointed out by Wallace in his work on lime sulfur. He points to the fact that the only method which approximates natural conditions is where the fungicide is allowed to dry before it is brought in contact with the spores.

The method developed by Wallace^o and Reddick^k is the best means of making tests of fungicides in the laboratory and comes very close to conditions in nature. This was the method on which the following tests were based with slight alterations in the method.

Wallace's method consists of spraying one-half of a glass slide with the spray to be tested, leaving the other half blank to serve as a check. The slides are allowed to dry under atmospheric conditions for some time and are then ready for the tests. In making these a suspension of the spores is made and a drop placed on each end of the slide.

These are kept moist and when sufficient time has elapsed the drops are examined and the percentage of germination on each end determined by count under the microscope. The comparison of germination on the check and on the spray will give a fair idea of the value of the fungicide.

This method was followed in the main in this work, a few minor alterations being made, which were as follows:-

^o Wallace, E. Cornell Bul. 290, 1911.

Seven cover glasses were sprayed with each dilution of fungicide used, these were allowed to dry at room temperature. These cover glasses were then used as tops to Van Tieghem cells, a drop of the spore suspension being placed on each one.

A separate check was not run for each drop but a sufficient number were made to be sure of an accurate determination of the average germination.

The spraying was done with an atomizer, taking care not to have too large drops and to cover the slide thoroughly. The spores were from a fresh culture on carrot.

The use of Van Tieghem cells instead of slides was to insure against drying out and to enable the spores to be counted with a 4 mm. objective, which is necessary in the case of small spores.

The fungicides were used while fresh and mixed in large enough quantities to insure that they were like those used in the field.

In counting, six fields of the microscope were used on each drop, all spores being counted in each one, thus getting a fair average of the whole.

The formula for each spray is given with the results of the tests in the following pages.

CHECKS.

A set of seven check slides was used for each series of spray tests conducted. The spores were germinated in distilled water on clean glass slips and careful count made to determine the actual average germination.

3. 4. 5. 6. 7.

Slide.	No. Spores Counted.	No. Germinated.	% Germination.
1	57	54	94
2	64	58	90
3	56	52	85
4	62	56	90
5	56	52	96
6	68	65	96
7	72	69	96

Average germination of check - - 92 %.

Second Set of Tests.

1	49	42	81
2	63	54	85
3	57	50	75
4	69	53	76
5	58	50	86
6	75	63	84
7	52	48	92

Average germination of check - - 85 %.

TESTS WITH BORDEAUX MIXTURE.

The Bordeaux used in these tests was made up from two stock solutions, one of CaO and the other of CuSO_4 . These were both made up on a basis of 1 gm. to 50 cc. of water, which is the equivalent of a mixture containing 8 lbs. to 50 gallons of water.

The two stock solutions were diluted separately and then poured together, being shaken for two or three minutes to insure proper mixing of the chemicals.

The spray was not allowed to stand or settle but was placed in a clean atomizer and sprayed on the cover slips at once. These had been soaked in cleansing fluid and washed thoroughly in distilled water before use.

After they had been sprayed they were allowed to dry for at least six hours at room temperature, to allow for any chemical changes due to atmospheric action, such as might occur when the spray was used in the field.

Two separate tests were made with Bordeaux mixture at an interval of some time, a new set of stock solutions being used and fresh cultures of the fungus. This was done to insure against error in the previous work and give a larger number of counts on which to basis the average amount of control secured by the use of different strengths of spray.

BORDEAUX MIXTURE.

4 - 4 - 50.

Stock solution of CaO - - 20 cc.

Stock solution of CuSO_4 - -20 cc.

This gives a mixture equivalent to a solution containing 4 lbs. of lime, 4 lbs. of copper sulfate and 50 gallons of water.

Slide.	No. of Spores Counted.	No. Germinated.	% Germination.
1	56	0	0
2	62	0	0
3	72	0	0
4	51	0	0
5	56	0	0
6	63	0	0
7	67	0	0
Second set of tests.			
1	46	0	0
2	52	0	0
3	58	0	0
4	61	0	0
5	47	0	0
6	63	0	0
7	59	0	0

4 - 4 - 50 Bordeaux in these tests absolutely control the fungus, no germinating spores being observed on any slide.

BORDEAUX MIXTURE.

4 - 2 - 50.

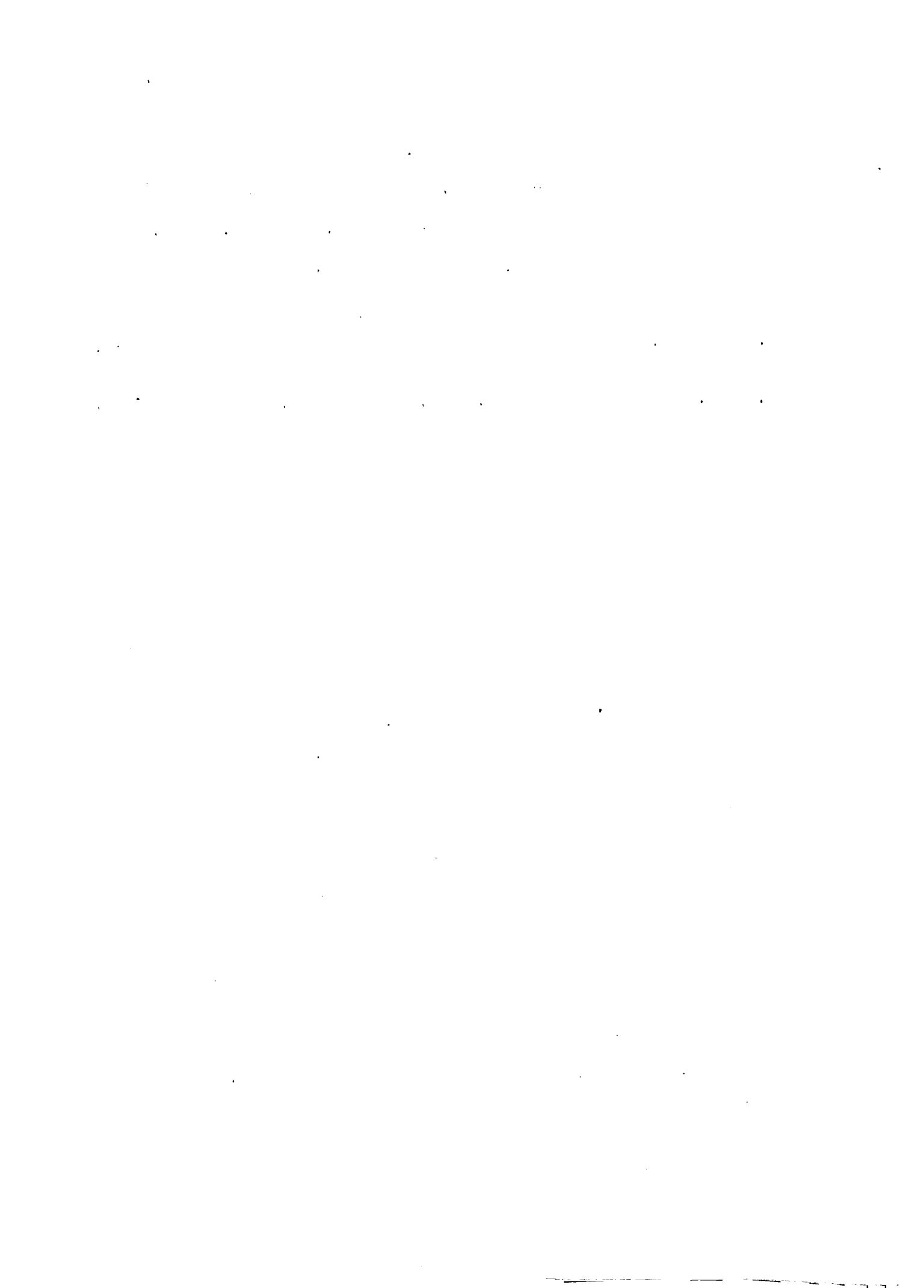
Stock solution of CaO - - 10cc. & 10 cc.water.

Stock solution of CuSO₄ - 20 cc.

This gives a mixture equivalent to one containing
2 lbs. of lime, 4 lbs of copper sulfate and 50 gallons of Water.

Slide.	No. of Spores Counted.	No. Germinated.	% Germination.
1	61	0	0
2	56	0	0
3	70	0	0
4	59	0	0
5	67	0	0
6	71	0	0
7	64	0	0
Second set of tests.			
1	58	0	0
2	64	0	0
3	57	0	0
4	69	0	0
5	72	0	0
6	49	0	0
7	81	0	0

4 - 2 - 50 Bordeaux in these tests gave absolute
control of the fungus, no germination being observed.



BORDEAUX MIXTURE.

4 - 3 - 50.

Stock solution of $\text{Ca}(\text{OH})_2$ - 30 cc. & 10cc. water.Stock solution of CuSO_4 - - 40 cc.

This gives a mixture equivalent to one containing
4 lbs. of copper sulfate, 3 lbs. of lime and 50 gallons of water.

Slide.	No. Spores Counted.	No. Germinated.	% Germination.
1	71	0	0
2	65	0	0
3	73	0	0
4	57	0	0
5	73	0	0
6	51	0	0
7	64	0	0
Second set of tests.			
1	64	0	0
2	67	0	0
3	83	0	0
4	74	0	0
5	62	0	0
6	70	0	0
7	65	0	0

4 - 3 - 50 Bordeaux in these tests showed perfect control of the fungus, no germination being noted.

BORDEAUX MIXTURE.

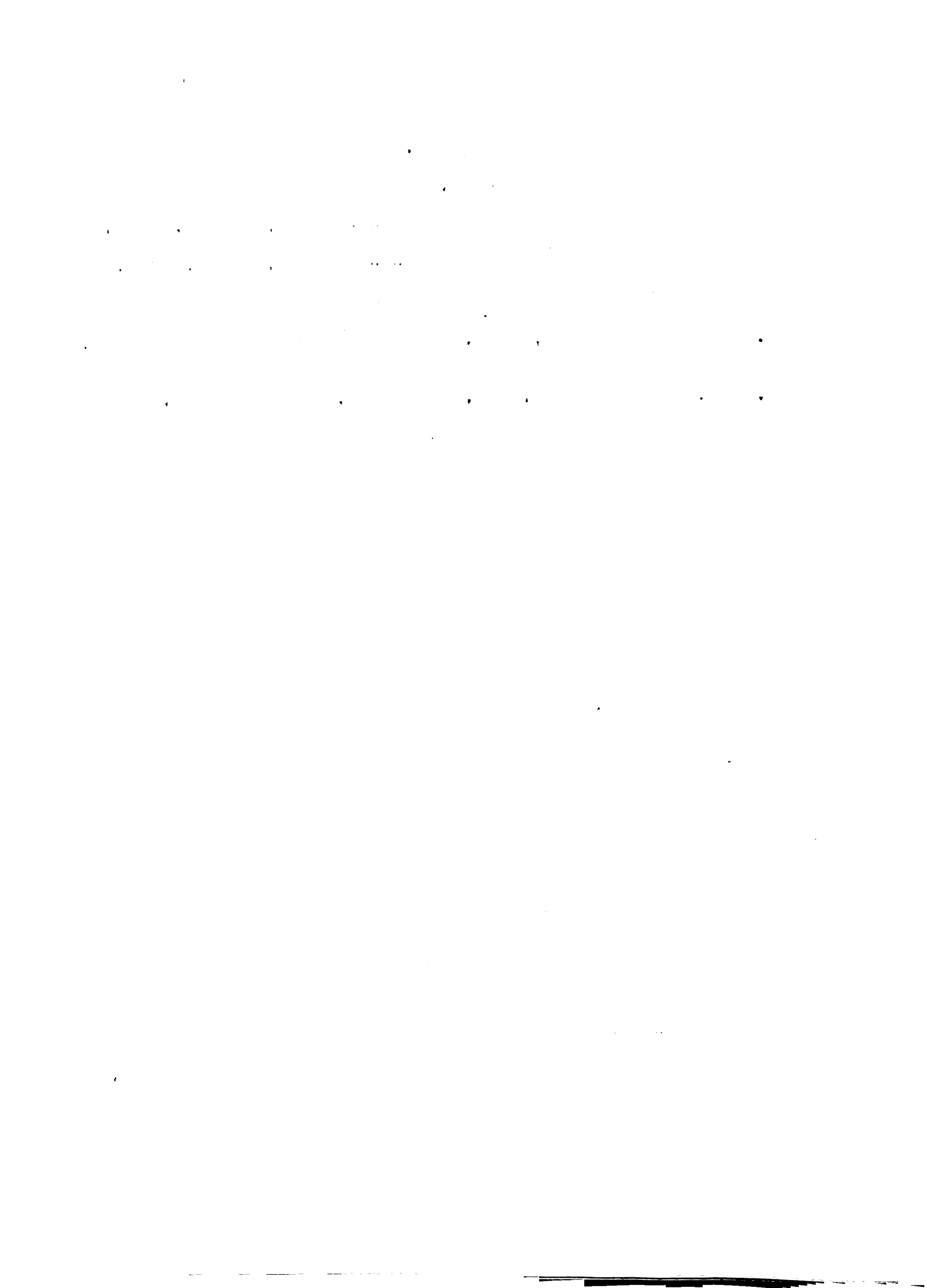
3 - 3 - 50.

Stock solution of $\text{Ca}(\text{OH})_2$ - - 30 cc. & 10 cc. water.Stock solution of CuSO_4 - - - 30 cc. & 10 cc. water.

This gives a mixture equivalent to one containing
3 lbs. of copper sulfate, 3 lbs. of lime and 50 gallons of water.

Slide.	No. Spores Counted.	No. Germinated.	% Germinated.
1	50	0	0
2	67	0	0
3	78	0	0
4	69	0	0
5	57	0	0
6	82	0	0
7	72	0	0
Second set of tests.			
1	59	0	0
2	64	0	0
3	51	0	0
4	68	0	0
5	73	0	0
6	55	0	0
7	54	0	0

3 - 3 - 50 Bordeaux showed no germination on any
slide which indicates that it is effective against the fungus.



BORDEAUX MIXTURE.

3 - 2 - 50.

Stock solution of $\text{Ca}(\text{OH})_2$ - -20 cc.&20cc.Water.Stock solution of CuSO_4 - - -30 cc.&10cc.water.

This gives a mixture equivalent to one having
3 lbs. of copper sulfate, 2 lbs. of lime and 50 gals. of water.

Slide.	No. Spores Counted.	No. Germinated.	% Germinated.
1	69	0	0
2	81	0	0
3	74	0	0
4	59	0	0
5	87	0	0
6	89	0	0
7	72	0	0

Second set of tests.

1	70	0	0
2	68	0	0
3	74	0	0
4	86	0	0
5	50	0	0
6	71	0	0
7	63	0	0

The fact that no germinating spores were found on slide in the tests of this mixture shows that it should be effective.

BORDEAUX MIXTURE.

2 - 2 - 50.

Stock solution of $\text{Ca}(\text{OH})_2$ - - 20 cc. & 20 cc. water.Stock solution of CuSO_4 - - 20 cc. & 20 cc. water.

This gives a mixture equivalent to one having
2 lbs. copper sulfate, 2 lbs. lime and 50 gals. water.

Slide.	No.	Spores Counted.	No. Germinated.	% Germinated.
1		50	0	0
2		48	0	0
3		56	0	0
4		69	0	0
5		46	0	0
6		71	0	0
7		63	0	0
Second set of tests.				
1		72	0	0
2		81	0	0
3		74	0	0
4		68	0	0
5		73	0	0
6		87	0	0
7		73	0	0

Even so weak a mixture as a 2 - 2 - 50, seems to
sufficient fungicidal power to prevent germination.

BARIUM POLYSULFIDE SOLUTION.

This is a proprietary compound, recently patented and not yet put on the market. Some was sent for experimental purposes and was tested on the fungus although it was unknown whether it would be possible to use it on the plant without injury.

The mixture was made up in three strengths as shown below. The solution was made readily by simply allowing the required quantity to dissolve in cold water.

Tests With 1 - 50 Solution.

Slide.	No. Spores Counted.	No. Germinated.	% Germination.
1	47	44	94
2	59	50	85
3	46	40	87
4	78	70	89
5	64	60	94
6	72	68	94
7	49	43	86

Average germination for tests with this strength of spray was found by the above table to be 89 %.

Since the checks run with these experiments show a germination of only 92 %, it is apparent that the dilution used would not be likely to prove effective.

BARIUM POLYSULFIDE TESTS.

1 - 30 Dilution.

This dilution was made by taking 1 gr. of the material and allowing it to dissolve in 30 cc. of cold water.

Slide.	No. Spores Counted.	No. Germinated.	% Germination.
1	71	55	77
2	62	42	67
3	51	33	64
4	63	51	81
5	72	54	75
6	64	52	81
7	58	42	73

Average germination of spores in this test was 77 %, which is somewhat higher than that which would represent valuable fungicidal action. Judging by the action of other sulfur fungicides on cucumbers it is unlikely that this could be used successfully on such tender plants in this dilution.

However even were it possible to use the compound, none of the dilutions used gave any indications that it would be of value in controlling the fungus attack. To be effective against so severe an organism as this has been proven to be, it would be necessary to have some fungicide which practically prohibit germination, as Bordeaux mixture seems to do.



BARIUM POLYSULFIDE TESTS.

1 - 40 dilution.

This dilution was made up by taking 1 gm. of the material and allowing it to dissolve in cold water, 40 cc. being used to give the required dilution.

Slide.	No. Spores Counted.	No. Germinated.	% Germinated.
1	80	75	93
2	64	58	91
3	73	64	87
4	87	76	86
5	57	50	87
6	59	53	89
7	63	57	91

Average germination for the tests with a 1 - 40 dilution of this spray 89 % which indicates very little fungicidal property for this dilution, in the case of *Cladosporium cucumerinum*, since the checks show a germination of only 92 %.

One effect of this chemical was very noticeable, however, that being the delaying of germination. All the spores were examined at the end of 36 hours and at this time most of those which germinated at all had developed a good sized germ tube. In the case of the spores placed on the cover glasses sprayed with this solution however, the germination was noted well begun before 40 hours although it proceeded at a normal rate after that time.

TEST WITH POTASSIUM SULFID.

This was used in a dilution corresponding to a common formula which is as follows:-

3 ounces K_2S .

10 gallons of water.

This is the equivalent of a mixture made with 1 gm. of K_2S and 427 cc. of water and the dilution was prepared by this formula from chemically pure K_2S .

Slide. No.	Spores Counted.	No. Germinated.	% Germination.
1	68	60	92
2	79	74	93
3	59	53	89
4	62	55	88
5	77	69	88
6	83	78	94
7	65	61	94

The average germination for the entire series was 91 % which shows that this spray would be useless as a control for scab, there being practically no control if any, since the checks showed 92 % germination.

The spores seemed to germinate slightly more rapidly in this solution than in water and conidia were even produced, which is unusual in water cultures.

TESTS WITH AMMONIACAL COPPER CARBONATE.

This is a fungicide that is much used where a non-staining compound is sought, but has not as strong fungicidal properties as Bordeaux mixture.

The common formula used is as follows:-

5 ounces Copper Carbonate.

3 pints Ammonia.

50 gallons of water.

This is equivalent to the mixture used in these tests, which was made up in these proportions:-

1gm. CuCO_3 .

8 cc. Ammonia.

1600 cc. water. Dissolve the copper carbonate in the ammonia and add water.

Slide.	No.	Spores. Counted.	No. Germinated.	% Germination.
1		58	48	85
2		72	63	88
3		81	76	94
4		64	56	82
5		67	55	82
6		78	62	79
7		51	46	90

The average germination for this set was 85 %, which does not represent much fungicidal value in the case of *Cladosporium cucumerinum*. This figure does not represent the true facts however as will be mentioned later.

TESTS WITH AMMONIACAL COPPER CARBONATE.

Second Series.

The chemicals were mixed in the same amounts and in the same manner as in the first set and all conditions were identical.

Slides.	No. Spores Counted.	No. Germinated.	% Germination.
1	74	60	81
2	58	41	71
3	69	54	78
4	63	59	93
5	79	68	81
6	55	42	79
7	67	51	71

The average germination for this series was 79 %. That for the whole series was 82 %. While as said before, this does not in itself seem to show fungicidal value, yet it was found that after germination the germ tube ^{was} ~~was~~ killed before growth had hardly begun and hence the fungicide proved to be effective though slow in its action.

It is such cases as these that makes Wallace's and Reddick's method so superior to that of merely dipping the spores into the liquid fungicide and then testing their powers of germination without having the germ tube come in actual contact with the fungicide after germination.



TESTS WITH DRY BORDEAUX.

This is a patented chemical compound and was used according to directions. A dilution of 10 lbs, to 50 gallons of water is recommended and it was made up by dissolving 2 gm. of the dry material in 78 ccc. of water, this giving the equivalent of the dilution recommended.

Slide.	No.	Spores Counted.	No. Germinated.	% Germination.
	1	47	0	0
	2	53	0	0
	3	80	0	0
	4	69	0	0
	5	74	0	0
	6	56	0	0
	7	54	0	0
Second series.				
	1	60	0	0
	2	54	0	0
	3	73	0	0
	4	69	0	0
	5	66	0	0
	6	78	0	0
	7	52	0	0

The dry Bordeaux mixture seemed perfectly effective in its fungicidal action, as no germination was noted in the set.



TESTS WITH ATOMIC SULFUR.

This is another proprietary compound and is recommended for use in a dilution of 5 lbs. to 50 gallons of water. The mixture as used in the tests was made as follows and is equivalent to the dilution recommended:-

1 gm. Atomic sulfur.

80 cc. water.

Slides. No.	Spores Counted.	No. Germinated.	% Germinated.
1	67	1	1
2	75	2	3
3	54	0	0
4	69	0	0
5	76	2	3
6	54	0	0
7	58	1	1

The average germination in the case of this spray was about 1 %, showing that if it is possible to use it on the foliage without injury, it might prove effective.

However the writer cannot say as to the danger of its use on cucumbers.

TESTS WITH POTASSIUM DICHROMATE.

This chemical was used merely as an experiment, without any idea as to whether it would be effective or whether it could be used on the plant. This was due to the fact that the work was all done on the presumption that Bordeaux mixture was ineffective and every possible fungicidal preparation was tried in an effort to secure something which would kill the spores of the fungus.

The first set of cover slips used with this chemical were sprayed with a .5 % solution of it in water.

Slide.	No.	Spores Counted.	No. Germinated.	% Germinated.
1	64	74	3	4
2		64	1	1
3		68	1	1
4		57	2	3
5		82	5	6
6		77	3	4
7		61	4	6

The average germination in this case was about 4 %, which would mean that it would prove an effective agent of control if the vines could stand an application of the chemical. Since Bordeaux, contrary to expectation proved that it is very strongly fungicidal to *Cladosporium cucumerinum*, however, there is no reason to pay much attention to the above results.

TESTS WITH POTASSIUM DICHROMATE.

In this set the dilution was made stonger, being 1 % instead of .5 % as before.

Second set .

Slide.	No.	Spores Counted.	No. Germinated.	% Germinated.
1		85	0	0
2		76	2	2
3		69	0	0
4		71	3	4
5		82	1	1
6		68	0	0
7		75	2	2

The average germination in this case was slightly over 1 % showing that this would effectually kill the spores of the fungus if it did not injure the plant too severely at the same time.

There was little difference in amount of germination between the two dilutions and if one were used, the .5% would be as valuable as the other.

This spray is so soluble in water however, that some other ingredients would have to be added to give it more adhesiveness, since otherwise it wash off in the first light rain or 1 heavy dews.

SUMMARY OF CONTROL MEASURES POSSIBLE,

The problem of controlling *Cladosporium cucumerinum* lies not in developing an efficient fungicide, for we have that in Bordeaux mixture as was shown by the tests described before. This spray, contrary to expectation proved very satisfactory in preventing germination.

It is the fact that the fungus fruits so rapidly and can be spread so easily that makes it a problem. The only way of combatting it seems to lie in the effective cooperation of the growers in the whole district.

The old vines should be burned each year, the cucumber crop used in rotation so as to avoid using land for pickles in two successive years and this rotation should be as long as possible.

In spraying there should be a concerted effort early in the season to prevent the fungus from getting a wide spread at the start. If every grower would spray carefully this might quite readily be accomplished but this is too much to hope for.

The spraying must be thoroughly done and if the time of spraying can be determined by weather indications as was discussed before, the results will be even more effective. By spraying as often as possible during August and adopting every possible measure in the way of cleaning out all infected material each season, much may be done to check this pest.

MEDIA.

Corn Meal Agar.

500 cc. water to which add 4 teaspoonfuls of corn meal and heat at 60° for 1 hr.

10 gm. agar added to 500 cc. water and boiled.

When heating is completed add to each solution enough water to restore to original volume and mix the two together.

Steam $\frac{1}{2}$ hr.

Filter, tube and autoclave 20 min. at 20 lbs.

Oat Agar.

50 gm. ground oats.

350 cc. water. Steam in cooker and then strain through cheesecloth. To this add 10 gr. agar dissolved in 150 cc. water and boiled thoroughly before adding.

Restore to original volume, steam, filter, tube and autoclave.

Cucumber Agar.

Grind cucumbers in chopper, press out juice, filter through cheesecloth. To this add 10 gm. of agar dissolved in 600 cc. of water.

Add egg albumen, steam for 1 hr., filter, tube and autoclave.

Nutrient Agar.

Dissolve 3 gm. extract of beef in 500 cc. water, add 10 gm. peptone, 5 gm. salt and to this add 500 cc. water in which has been dissolved 10 gm. of agar. Steam 30 min. add egg albumen, boil 10 min. over free flame, filter, tube and autoclave.

Hard Potato Agar.

250 gm. sliced potato.

1000 cc. water.

Cook $\frac{1}{2}$ hr. in steamer.

Decant, restore to original volume.

20 gm glucose.

30 - 50 gm. agar. Cook in steamer 3 hrs.

Use 1 egg to each liter to clear. Filter, tube and autoclave.

Potato plugs.

Wash potatoes thoroughly with a brush, soak in $HgCl_2$ for 10 min. and rinse in tap water.

Pare thick, wash over night in running water, rinse in distilled water. Place in sterile tubes containing moistened cotton in bottom

Steam 15 min. on three successive days, at 100° .

Carrot Plugs.

Prepare same as potato plugs.

Parsnip Plugs.

Prepare same as potato plugs.

Corn Meal.

3 gm. corn meal and 10 cc. distilled water placed in test tube and autoclaved 30 min.

Rice.

$2\frac{1}{2}$ gm. rice and 10 cc. water placed in a test tube and autoclaved 30 min.



Celery Stems.

Fresh celery stems were scrubbed in tap water, washed in HgCl_2 for 5 min. rinsed in tap water afterwards in distilled water.

Cut in 2 in. lengths, place in test tube containing cotton moistened in distilled water. Steam for 3 days, 15 min. each day at 100° .

Cucumber stems.

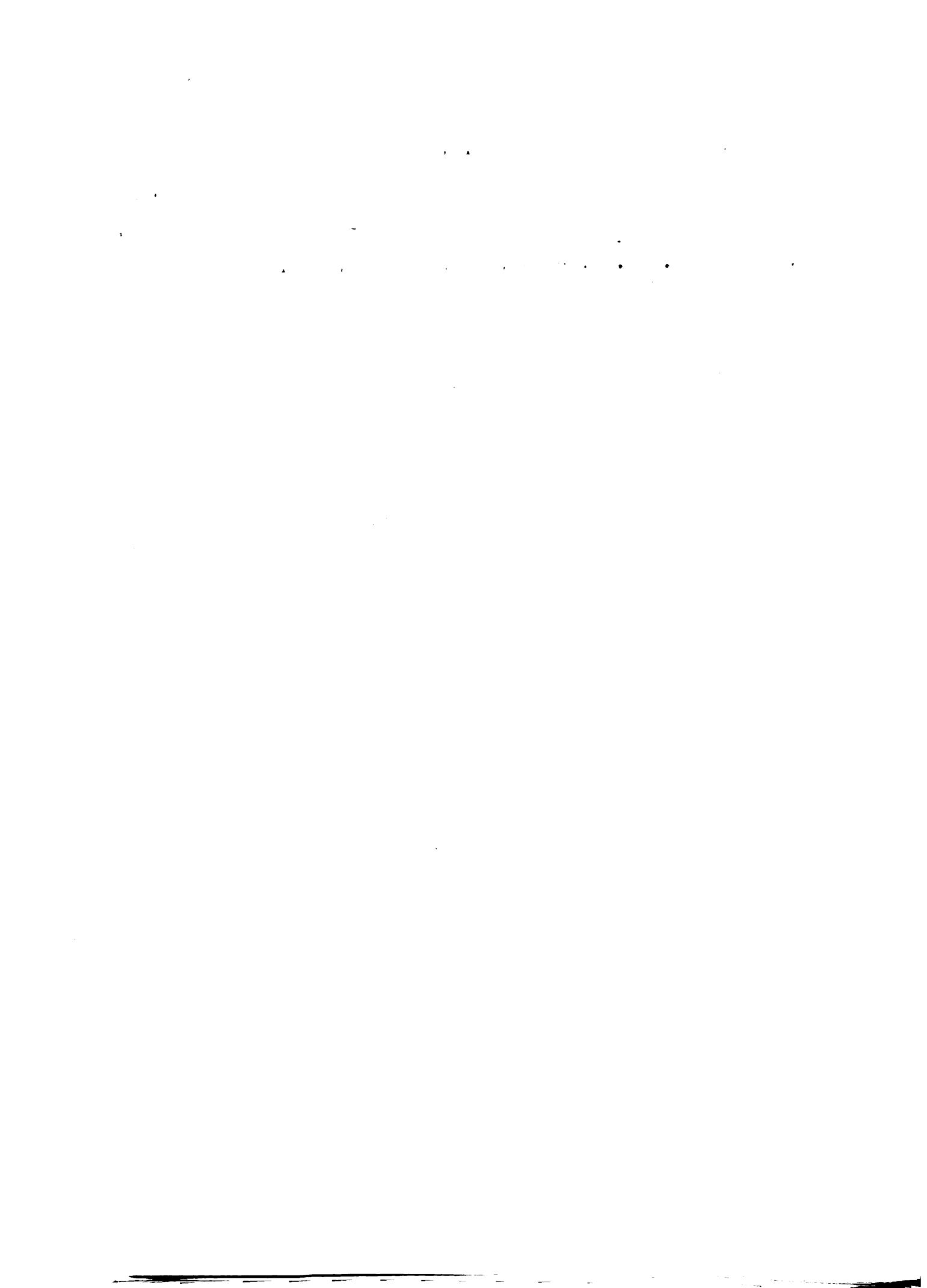
Prepared exactly in the same way as celery.

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EXPLANATION OF PLATE I.

- Figure 1. Conidia formed from spore 12 hours after germination.
- Figure 2. Manner of growth of conidia.
- Figure 3. Fruiting hypha, showing fusion of two strands of mycelium, only short portion shown.
- Figure 4. Fruiting hypha again showing fusion, not the same case as Fig. #.
- Figure 5. Swollen segments in mycelium on nutrient agar.

PLATE I.

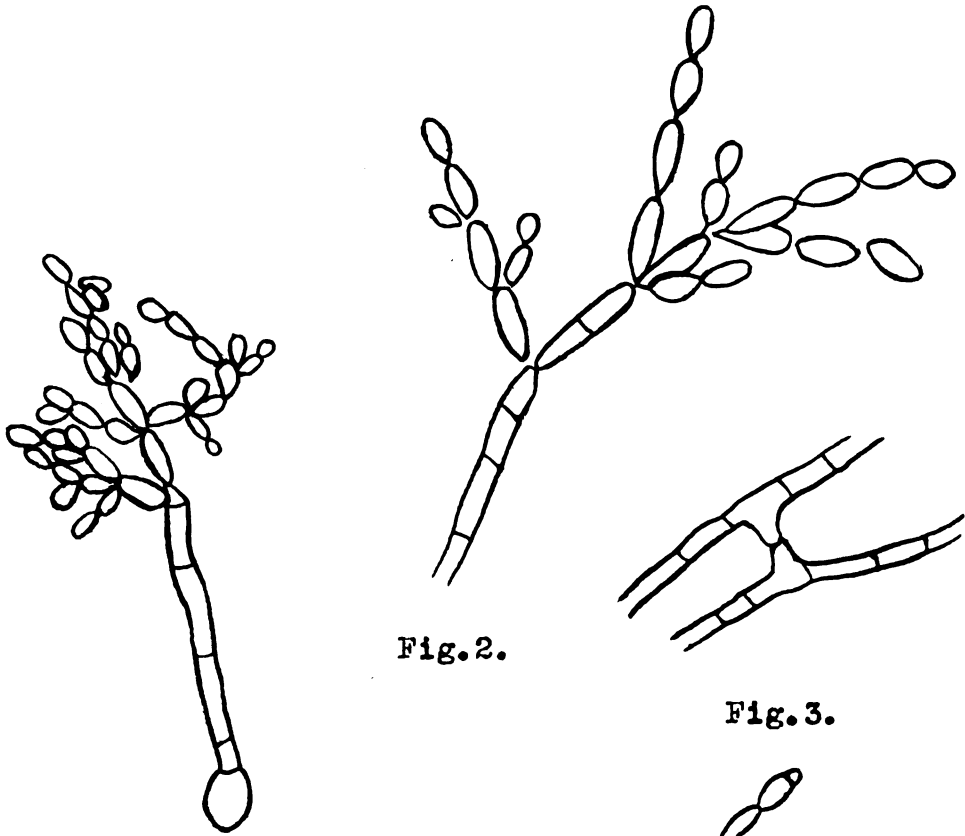


Fig. 1.

Fig. 2.

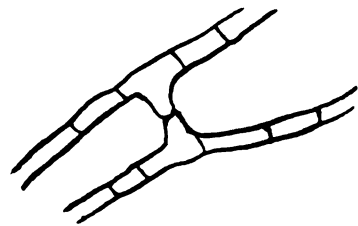


Fig. 3.

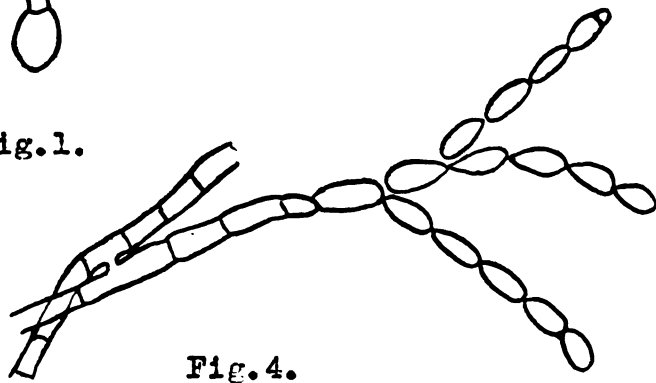


Fig. 4.

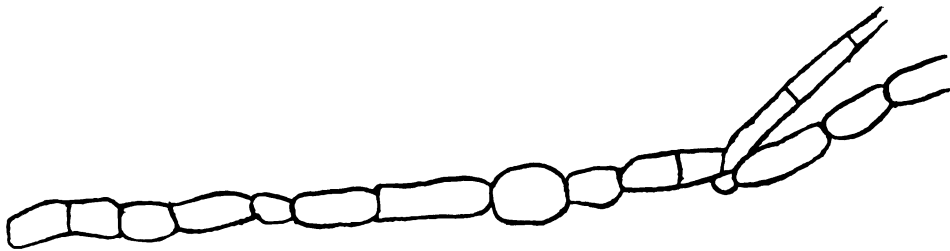


Fig. 5.

EXPLANATION OF PLATE II.

Figure 1. Spore of the fungus.

Figure 2. Spore 20 hours after being placed in water.

Figure 3. Same spore 3 hours later.

Figure 4. Same spore 1 hour after previous drawing.

Figure 5. The spore at the end of 30 hours.

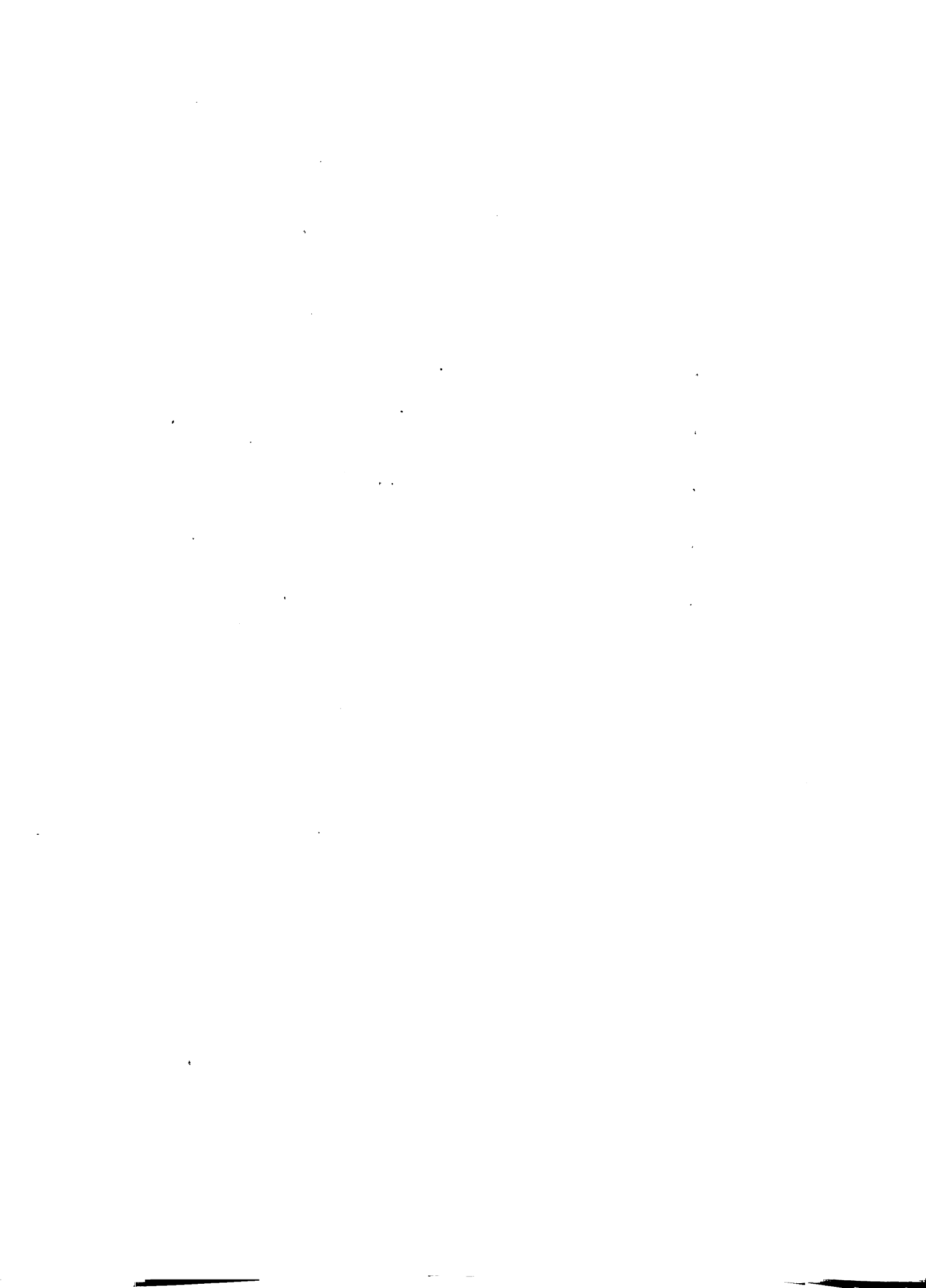


PLATE II.



Fig. 1.



Fig. 2.



Fig. 3.



Fig. 4.

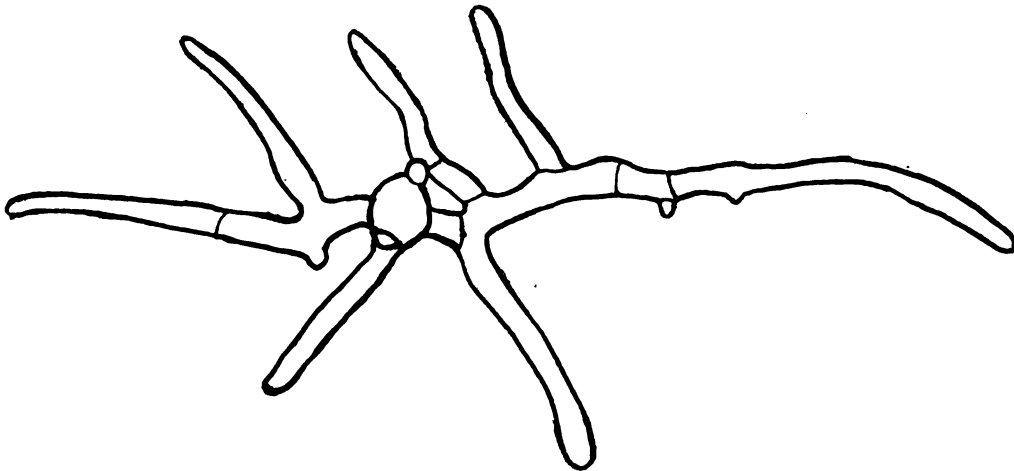
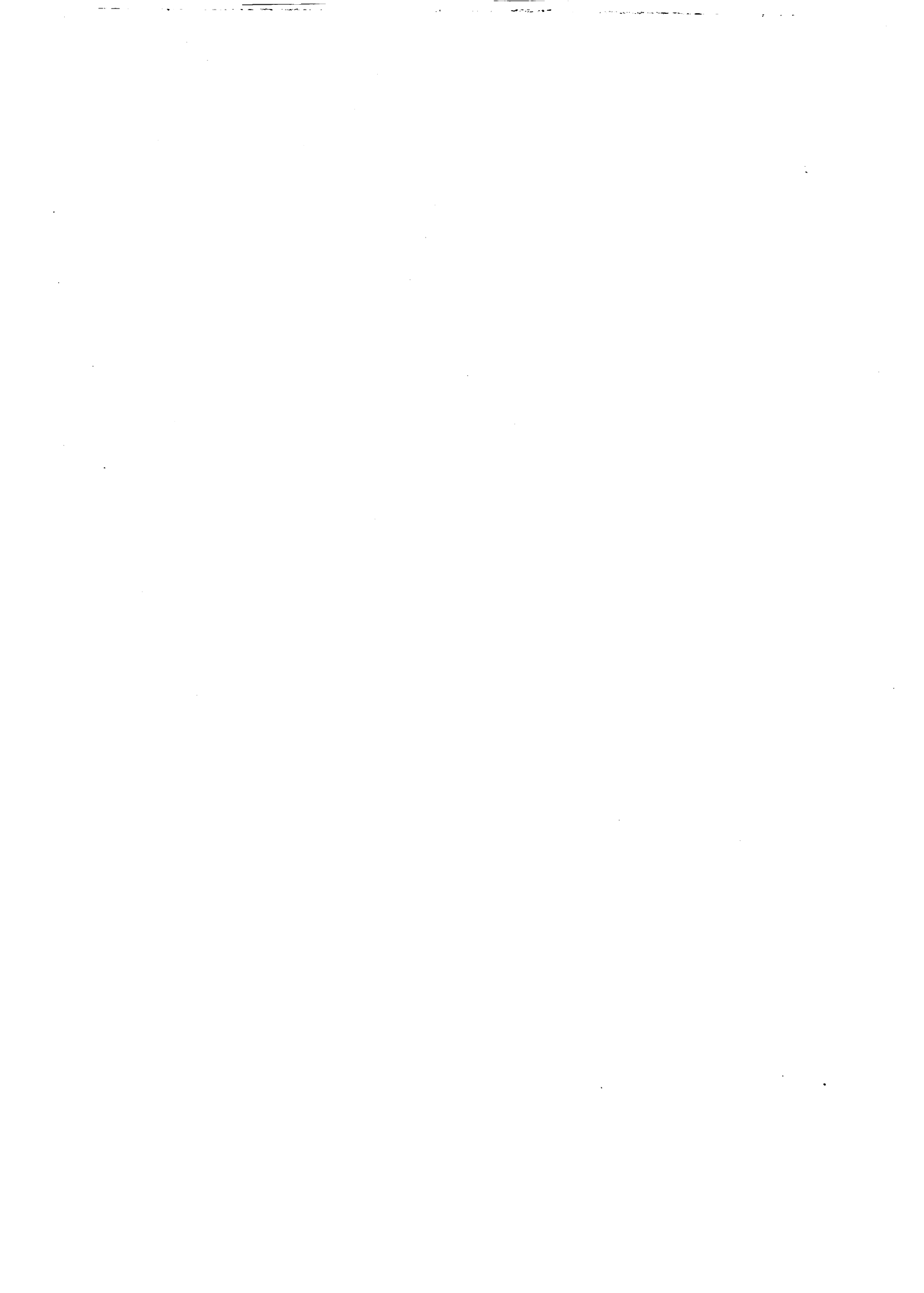


Fig. 5.





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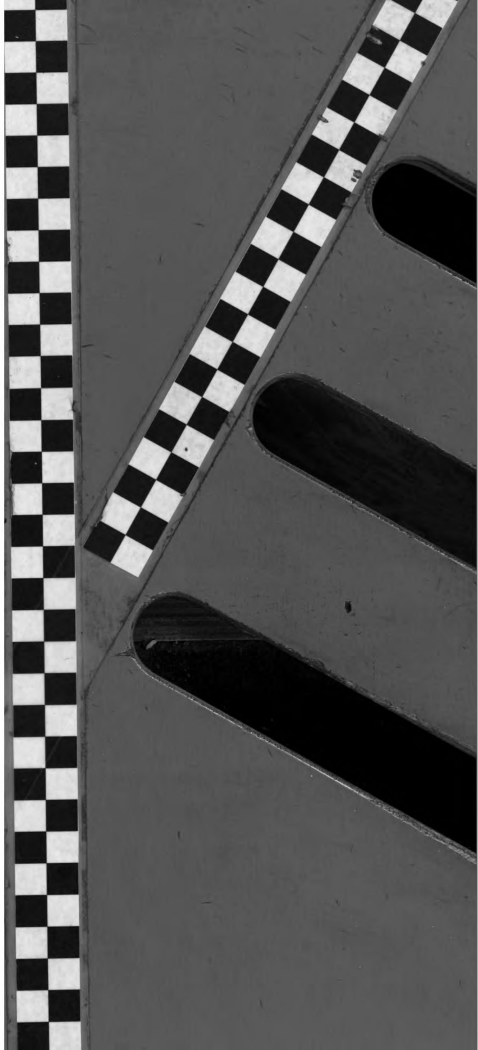




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