STUDIES ON THE SUNFLOWER RUST
PUCCINIA HELIANTHI SCHW.

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Studies on the Sunflower Rust, *Puccinia helianthi* Schw.

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Introduction.

The sunflower, long playing an important part in the agriculture of Northern Europe, has become recently a crop of great promise for the Northern part of the United States and Southern Canada. Widespread interest is being shown in the crop and the acreage is increasing rapidly. Its usefulness, however, may be somewhat impaired by a disease which sometimes reaches the proportions of a serious epidemic. The purpose of this paper is to bring out some little known or doubtful points and some new facts concerning this disease, the sunflower rust.

History of the Sunflower (Helianthus annus Schw.)

Helianthus annuus is recognized as a native of North America with a natural range of distribution extending southward to Peru. Palmer, in his article on the food products of the South American Indians, says the seeds of sunflower were often gathered and "being very sweet and oily they are eaten raw or pounded up with other substances made into flat cakes and dried in the sun, in which form they appear to be very palatable to the Indians". Champlain in 1615 found Indians of Georgian Bay cultivating the sunflower and using the oil obtained on their hair.

The earliest record of the sunflower appears to be in a history of flowers by Rembert Dodoens (15) which was published in 1568 at Anvers, France. Dodoens described and figured Helianthus annuus here for the first time under the name of Peruvian Chrysanthemum as one of the new and curious plants to be found in the Royal Gardens. He received the rare plant from a friend in Spain and he states that the plant is reported to have been found in Peru and in several other provinces of America. About this same time the plant was introduced into Italy where it grew to enormous stature, if the reports of Jacob Antoine Cortusus, a noted physician, are to be believed, attaining a height of forty feet. Cortusus (15) reported, after a successful trial, that "the leaves are very tasty when scraped to remove the hair, cooked on a grid iron, and prepared with salt and oil, also the flower receptacles similarly prepared are more delicate than the bracts of the artichoke." Monardes (15), a doctor of Seville a little after Dodoens described the sunflower and recommended it to embellish gardens and showed that the plant was getting fairly common among the collectors of plants in the gardens of France. Charles de l'Escluse in his work (14) on the Exotics (1605) described the sunflower and stated that for several years it had been commonly known by all Europe. He distinguishes two species, the one unbranched and single flowered, while the other was much branched and many flowered. Fragosus de Toledo, a Spanish botanist, gave a very complete description of the sunflower in his Rapsodies (17). In the

Universal History of Plants by Jean Bauhin (7) is found a resume of the observations on the sunflower by the various writers at the end of the sixteenth and the beginning of the seventeenth century. Jean Bauhin called it <u>Herba maxima</u>, the largest herb, and adds that Lobel gave it the name of <u>Gigante</u> in Portugal. He accepts with Cesalpin the name <u>Flos Solis</u>. Gaspard Bauhin, in his Phytopinax, remarked on the variability of the plant in size of flowers, its leaves, the height of its stalk, and the number of its flowers (6). He observed that the flower is sometimes white.

Jean Bauhin in a later article added that he grew both the small and the large variety, and mentions also another very small variety which had a multitude of flowers.

We find the sunflower entered by Guy de la Brosse in 1636 in his "Catalogue of Cultivated Plants in the Garden of the King" under the name of Peruvian Chrysanthemum or <u>Flos Solis</u>, and also a <u>Chrysanthemum Peruviarum proliferum</u> which appears to be <u>Helianthus multiflorus</u> L.

In their Amateurs' Manual of the Garden (12), Decaisne and Naudin state the Great Sunflower or <u>Tournesol</u> is somewhat naturalized in Europe and from it have arisen several varieties which reproduce their seed, among others is mentioned the giant variety, the double varieties, etc.

From France the sunflower spread northward as it became acclimatized (for the first sunflowers brought into

France would not mature their seed there) until it was first grown in Russia in the Province of Woronesh 1830-1840, authorities differing on the exact date. They were first grown on a large scale, according to Woronin (33) in a peasant's garden in that province in 1841, the peasant pressing their seeds for oil. This was the start of the great sunflower oil industry which began to make the farmers of the region wealthy as the demand for the oil grew and the price increased.

Economic Importance of the Sunflower (*Helianthus annuus* Schw.)

Uses of the Sunflower-

Sunflowers have been cultivated for many purposes since the Indians were found cultivating them for their seed and oil early in the seventeenth century. They have been widely used as an ornamental, while the seeds are a valuable food for birds and poultry and are used by horsemen as a remedy for heaves. In Europe the table oil was made by cold pressing the seeds and is in demand, and the seeds themselves are eaten raw or roasted like the peanut in this country. The oil, states the Scientific American (1) that is not suitable for edible purposes is used for burning, for soap-making, and for an inferior varnish oil. Immense quantities of oil cake are produced in Russia and shipped to Denmark where it is popular as cattle food. The stems may be used as a source of potash for the ash of the stems contains

39% K_2O and the seed husk 56%. An acre of stems will give from 40 to 50 pounds of potash, according to the Scientific American. The pith of the sunflower stalk has long been used by French surgeons as a moxa and it is now used extensively as a substitute for other materials formerly employed in making these moxas for cauterizing purposes. In 1915 a new utilization of the sunflower pith became generally known through another Scientific American article (4) on "The Lightest Known Vegetable Substance". The pith of the sunflower stalk is this lightest known vegetable substance with a specific gravity of 0.028, while the pith of the elder is 0.09 and cork 0.24. This discovery has essentially increased the commercial value of the plant. The pith is now carefully removed from the stalk and applied to many important uses. One of its chief uses is in the making of life saving appliances. Cork with a buoyancy of one to five, and reindeer's hair with one of one to ten have been used; the pith of the sunflower has a buoyancy of one to thirty-five. This new material is being used in the construction of boats and life preservers. A sufficient amount can be worn on the person without any inconvenience.

Importance in America-

There are three important seed producing areas in the United States. These are southeastern Missouri, southern Illinois, and the San Joaquin Valley of California. The

United States Department of Agriculture Market Reporter (2) estimated the 1920 crop in these three areas to be 9,850,000 pounds.

The greatest importance of the sunflower in America, however, lies in its use for silage. United States Experiment Stations commenced to report experiments with sunflowers for silage in 1883 at New York, in 1893 at Vermont, and in 1895-1896 at Maine, while the first Canadian report was in 1893. Most of the investigators have obtained a larger yield of sunflowers (total crop) than they did of corn, but the early investigators did not ensilo the whole plant, having the idea that there was little food value in the stalks. Canadian authorities reported that butter made from the sunflower silage had a richer flavour and higher color than that from corn silage. Since these early tests, but little investigation has been made until recently through small trials were reported by New Hampshire, Nebraska, Colorado, and a few other states. In 1915 the Montana Station grew a small acreage under irrigation with such success that the planting was enlarged in 1916 and the preliminary tests described in Bulletin 118 of the Montana Station demonstrated the high feeding value of sunflower silage and resulted in a widespread interest in the crop. Numerous other states as well as the United States Department of Agriculture have since been experimenting extensively with it as a silage crop. Their value in any region depends on the success to be had in that region in growing other crops. Vinall, of

the United States Department of Agriculture, made an extensive study into the problem and says: "Sunflowers will not supplant corn in the corn belt nor sorghums in the central and southern Great Plains because these silage crops do so well in supplying the need for a silage crop under their own climatic conditions." "In the same way the sorghums and millets supply the southeastern states. However in the extreme northern part of the United States or at high altitudes in the West where the temperature during the growing season is low, corn, sorghum and the millets do not produce heavily and in these regions sunflowers are being recognized as an extremely valuable silage crop and the acreage is increasing rapidly." It is probable, says Vinall in his U. S. Bulletin on "Sunflower as a Silage Crop" (32), that sunflowers for silage will become widely grown in the New England states, northern New York, Michigan, Wisconsin, and Minnesota, in North Dakota, Montana, Washington, and Oregon, and also in some of the high valleys of the Rocky Mountain region such as the San Luis Valley of Colorado. Sunflowers are frost resistant so that they are green two or three weeks after corn has been killed by the frost. The crop has been successfully grown on sandy soil in northern Michigan and on poor clay soil in West Virginia while the best yields are made on rich clay loams with abundant humus. It is the coming forage crop north of the frost line where good yields of corn are not obtained.

One of the problems of this area north of the corn belt is to find a suitable cultivated crop to place in the rotation; another is to raise a sufficient tonnage of succulent feed to make the raising of cattle possible. The sunflower answers these problems and is rapidly increasing in acreage. It practically pushes the frost line 50 miles north and extends the cattle feeding and dairy industry above our northern boundary into southern Canada.

Importance in Europe-

In northern Europe the sunflower plays a very important part, especially in Russia, as one of the important crops of the region and rated by Karsin as equal in importance to the sugar beet. Oil production from the sunflower is on an enormous scale there. Data from the city of Saratov given by Karsin (19) shows that the first factory was founded in the forties. By 1888 there were 33 large factories, equipped with British and American presses, which produced over 28,800,000 pounds of oil. By 1893 the production reached 54,000,000 pounds of oil from 238,000,000 pounds of seed. The value of the 1894 crop was 7,382,050 roubles or over $3,800,000 at the pre-war rate of exchange, the farmers receiving about 17¢ per pound for their seed (1 rouble 17 kopecks per pood). In addition to the oil production, the oil cake production reached 66,000,000 pounds in that year from the city of Saratov. Practically

all of this oil cake was exported, giving employment to a considerable number of ships. Saratov leads in oil production so that these figures represent about 80% of the total Russian production. The production was considerably curtailed during this period, as willbe shown under the heading "Economic Importance of the Sunflower Rust" by the epidemics of rust.

Economic Importance of the Sunflower Rust.

The one factor which may partially offset the advantages and limit greatly the value of this new crop is the sunflower rust. The exceedingly strong parasitism of this fungus kills the lower leaves and in severe attacks all of the leaves. This causes a great loss of succulence and of the tonnage of silage. In the feeding experiment conducted at the Michigan Experiment Station in 1919 by Brown (3), the sunflowers were very severely rusted and the crop harvested so late that there were very few leaves still green upon the plants. Accordingly, in his experiments, the feeding value of sunflower silage made from these bare stalks was not as high as other investigators have found them to be.

In addition to the loss of succulence, there is a drain upon the plants from severe attacks of the rust which results in a poor filling of the heads and a consequent lowering of the fat content in the silage.

The most severe and most economically important attack of the rust fungus on the field sunflower occurred in Southern Russia in 1866-1868, in the province of Woronesh. At the time the epidemic struck the province the farmers were becoming wealthy from the great development of this comparatively new industry. Large fields of from eight to sixteen hundred acres were planted to sunflowers for oil-seed production. Then the farmers began to have crop failures due to the rust parasite and, says Woronin (33), "In 1867-1868 the disease took on such frightening proportions that it was necessary to throw away the harvest of entire fields and to substitute other crops----so that where sunflowers grew in areas of hundreds of acres in stretches of several miles, now in 1872 one meets with but an occasional small field in a twenty-five mile drive." The province never entirely recovered from this blow to the sunflower industry, for although Woronin made a careful study of the parasite and advocated some control measures, these were not put before the people in popular form and no control measures were adopted.

Thirty years later Karsin (19) writes that the sunflower had spread into the province of Saratov where it began to be cultivated on an enormous scale. For a few years the sunflower yielded well in Saratov, then the rust reached a serious state of epidemy in this province just as it had previously done in Woronesh. Karsin says in 1896 (19) that "during the previous ten years there were five poor crops, two medium crops and only three good crops".

History and Distribution of the Sunflower Rust.

A rust on sunflower was first observed and described by Schweinitz in 1822 in his Synopsis Fungorum of North Carolina. He described there the aecial stage as an Aecidium (27), the telial stage on Heliopsidis and Vernoniae as one species (28), and the telial stage on various species of Helianthus as another species (29). Sunflowers, due to their varied uses, are widespread in their distribution. "They are grown throughout North America from Canada to the Canal zone and in most of South America, but more especially along the west coast from Columbia to Chile. Sunflowers are also grown to a limited extent in Australia, New Zealand, South Africa, Egypt, the Mediterannean region of Europe, India and China", according to Vinal (32). Russia is the leading country in the world in the production of sunflowers being grown on a large scale for the seeds and oil. Next to Russia, Hungary is the largest producer of sunflowers. This widespread distribution of the sunflower makes the distribution of the rust very widespread also, for wherever the sunflower has gone the rust has appeared. Since Russia was the largest producer of sunflowers it was here the effect of the rust on the crop first began to come to the notice of the people. In 1866 the first complaints from the Russian farmers were heard regarding a rust epidemic and during the following two years the sunflower industry was dealt a very severe blow from which it never recovered. In the period 1890-1900 the rust epidemic reached the great sunflower plantings in the province of Saratov.

Signs of the Disease.

Spring-

The disease first becomes visible in the spring in the form of minute yellow pycnia on the cotyledons of the seedlings about two weeks after planting. These are usually followed in about five days by the development of clusters of aecial cups forming bright yellow spots which may appear on either surface of the cotyledon. It is not unusual to find several groups of pycnia or several clusters of the aecial cups on the cotyledons but relatively few plants are founds with their cotyledons so infected. Where volunteer seedlings come up in dense groups from a buried portion of a head, the infected seedlings may reach ten percent of the total in the clump. Very rarely in the field the yellow spots of the aecial cups are found on the true leaves of the young seedling. The comparatively small number of infections and their lack of conspicuousness makes it very easy to overlook this stage, though it is very important.

Summer-

The first stage, which is seen and recognized by the casuál observer, is the summer stage or uredinial stage. In 10 to 12 days after the appearance of the aecial cups the first reddish brown pustules of urediniospores appear. The lower leaves seem first attacked, gradually spreading upward and the lower surfaces of these basal leaves become nearly covered by the rusty brown pustules of the rust while occasional pustules appear on the upper surfaces of the leaves. The pustules appear as powdery, red brown masses from which a fine brown dust arises on touching the leaf. These increase rapidly in size and numbers, and in a few weeks the lower leaves are almost entirely covered with the brown pustules. These worst infested leaves wither and die and we find the leaves entirely dead up to a height of two or three feet by the latter part of August. Not only the leaf blades are marked by the brown spots, but pustules appear thickly crowded on the petioles, the main stem itself has scattered groups, and even the involucre and floral bracts become well spotted by the uredinial pustules, while occasionally the flowers themselves, especially the ray flowers, show rust spots. The development of the rust is especially rapid during the late summer through the month of August in this climate.

Fall-

At the end of August and beginning of September the powdery reddish brown rust pustules of the summer begin to darken in color and become more compact in form due to the formation of the fall spores or teliospores. As the proportion of teliospores becomes greater, the spots all become dark brown or brownish-black in color giving the characteristic fall appearance of the disease. Thus we have three characteristic signs of the disease as the season progresses,- the Spring stage, important but inconspicuous, usually manifests itself by yellow spots of pycnia and aecia on the cotyledons or on the first true leaves of the seedlings. The Summer stage is characterized by the rapid spread of the parasite showing itself by the reddish brown rust pustules which kill the lower leaves and seriously injure the others. The Fall stage is characterized by dark brown rust pustules showing very conspicuously on leaves, stems and involucre of the sunflower plant.

Life History of the Causal Organism.

Previous Work-

There has been more or less doubt and uncertainty as to the exact life history and identity of the Puccinia (Dicaeoma) which causes the rust of the common field sunflower Helianthus annuus.

Woronin (33) found the fungus causing the rust epidemics on sunflowers in Russia to be the Puccinia helianthi Schw., described by Schweinitz (28) in 1822. He found this autoecious rust had all of the five spore forms of a rust, namely - teliospores, sporidia, pycniospores, aeciospores and urediniospores.

The Sydows, however, in their Monographia Uredinearum (31) comment on Woronin's work as follows: "Woronin states that he has obtained aecidia through the sowing of teleutospores on Helianthus annuus. Neither before him nor since this time have aecidia been found on this Helianthus although the species occurs commonly in many European regions so that the statements of Woronin concerning the possession of an aecidium by the Puccinia are still doubtful. The commonly occurring aecidium of different species in North America has never been observed in connection with the widespread Puccinia occurring there, whence the aecidium may be an isolated form or belong to a heteroecious species. In our opinion, this species (Puccinia helianthi) possesses no aecidium."

Arthur (5), however, describes the aecial stage in his work on the Uredinales in North American Flora.

Methods of Study-

In studying the sunflower rust in its various stages the plants were inoculated and when the organism reached the proper stage of development small squares about 5 mm. square were cut out of the leaves and killed in Gilson's fluid. This fixer proved to be best adapted for the purpose since it has a stronger tendency to fasten the spores so that they are not washed off in the subsequent transfers in the process of dehydration and infiltration. The paraffin method of imbedding was used and the sections were cut from 2 to 10 microns in thickness. Due to the spongy nature of the young leaf tissue with its large intercellular spaces, 7 microns proved best in most cases. In studying the morphology of the pycnia and the aecial cups, sections were cut but 13 microns in thickness. The Durand method of staining (16) with Delafield's haematoxylon and alcoholic eosin for intercellular mycelium gave brilliant results for the mycelium itself. To bring out the cytological details of the fruiting bodies and the spores, Haidenhaimb iron alum haematoxylon (11) stain was found preferable since the spores take the eosin of the Durand stain too deeply.

But one investigator, J. Ray (24) has reported the successful growth of a rust in a culture medium. His method was followed and in ten trials gave negative results with this organism. Since it was apparently im-

possible to grow this fungus in pure culture in a synthetic medium, the growth of the parasite in pure culture was accomplished upon the living host. Seeds were sterilized by the method found best by Young (35) presoaking in water for 18 hours then dipped in 3% formaldehyde for 30 minutes. The fruits were then carefully opened and the kernel removed by sterile forceps and placed on nutrient agar poured in a thin layer in deep culture dishes. Four kernels were placed in each dish and in about 75% of the dishes all the seed remained sterile as shown by examination of the surrounding agar. Large test tubes 2 1/2 inches in diameter and 12 inches long were used for growing the plants. Shive's synthetic solution #52 was used as the most favorable liquid medium known for plant growth. Expressed as normal solutions this medium contains 18 cc. KH_2PO_4, 5.2 cc. $CaNO_3$, and 15 cc. of $MgSO_4$ made up to one liter with distilled water. Quartz sand saturated with this solution proved to be too compact for good growth of the seedlings. Best results were obtained by forming small supports of glass which held a small pad of absorbent cotton with the lower edge of the cotton in the solution. Sufficient aeration was provided for the roots in this way and the large tubes plugged with the glass supports and cotton in place were easily sterilized in the autoclave. Sterile seedlings from the deep culture dishes were then transferred to the large test tubes by flamed forceps. When twenty tubes contained sterile plants of from 6 to 8 inches high, inoculations were made. The top of a pustule of urediniospores from a plant in the greenhouse was cut off with a sterile scalpel and a few of the spores from the interior of the pustule transferred to a leaf of several of the sterile plants. Typical uredinia appeared in ten days and a second transfer was made by a sterile needle to other sterile plants. The uredinia which developed on these were assumed to be sterile, but more transfers were made from time to time thus continuing the growth of the rust in pure culture for five months. Pycniospores, aeciospores, urediniospores and teliospores were thus produced in pure culture and sections of leaves were fixed, imbedded and sectioned from time to time. From these sections, augmented by material not grown under sterile conditions, the life history and the morphology of the fungus was studied.

Identity of Species-

The modal curves on Plate III show the measurements of the spores from the Puccinia helianthi of Europe compared with the measurements of two American specimens of the Dicaeoma helianthi-mollis of Arthur. The almost exact similarity of these curves would seem to prove

Woronin's and Arthur's statements and descriptions are of the same organism, the Puccinia helianthi of Schweinitz. It is evident that Sydow was mistaken when he stated the Puccinia helianthi of North America had no aecial stage. The plates numbering IV, V, and VI show the aecia on Helianthus annuus obtained by inoculating with teliospores of Puccinia helianthi from Helianthus annuus of the previous year. The entire life history as determined by numerous inoculations using both the teliospores of the Helianthus annuus and those from other wild forms of Helianthus such as Helianthus tuberosus, Helianthus giganteus, and Helianthus multiflorus follows.

Life History-

The teliospores which have wintered over as pustules on old leaves and stems germinate very easily in the spring, forming promycelia. This promycelium soon divides into four cells, each of which extrudes a prolongation which rounds up at the end and swells up to form a spherical or oval sporidium into which the contents of the cell pass. The sporidia are then cut off and projected some little distance by the promycelia. When these sporidia alight on the cotyledons or tender young leaves of the sunflower seedling they germinate by means of germ tubes if enough moisture is present. These germ tubes enter the stomata and soon develop into a considerable mass of one-nucleate mycelium. Ten days after the germination of the sporidia the mycelium develops pycnia on the upper surface of the leaf. Five days later aecial cups are usually formed, either on the same or the opposite side of the leaf. Occasionally there is found to be an elision of the aecial stage. This appears to be caused by dry conditions after the mycelium has commenced to grow in the leaf tissue. When this elision of the aecial stage occurs instead of finding aecial cups developing following the pycnia, uredinia may be observed developing with no external evidence of the aecial stage. Under the normal damp conditions of early spring the aecial stage develops and the aeciospores are carried by the wind to other leaves of the sunflower. There they germinate in a drop of moisture usually by two germ tubes, one of which will enter a stoma and start the development of the two-nucleate mycelium. From this two-nucleate mycelium we have the urediniospores developing in about ten days. The uredinial stage repeats itself every ten days during the summer. In the fall the brown uredinial pustules darken in color due to the formation from the mycelium of the dark brown teliospores among the urediniospores and as the season progresses the pustules consist entirely of teliospores. These teliospores winter over and germinate

commencing the life cycle again.

Morphology.

Mycelium-

Puccinia helianthi has a much branched, septate mycelium varying from 1 1/2 to 5μ in the diameter of the hyphae. At first wholly intercellular with haustoria extending into the air spaces of the mesophyll, later intracellular in the areas near the fruiting bodies. Stains brilliantly with the Durand stain (16) for intercellular mycelium. The hyphae appear more or less granular and contain many fine vacuoles.

Pycnia-

The pycnia are amphigenous on the cotyledons but are usually epiphytous on the true leaves in close groups 1-2 mm. in diameter. Their color is a light yellow with a waxy luster, becoming a dirty yellow with age. The pycnia are globoid about 100-160μ in diameter. The ostiolar filaments are agglutinated into a column 70 to 120μ long. The pycniospores are spherical or oval 1 1/2 - 2 x 2μ in diameter.

<u>Aecia</u>-

The aecia are amphigenous on the cotyledons but are usually hypogenous on the true leaves. They are collected in small groups 1-4 mm. in diameter on the true leaves but usually smaller, 1-2 mm, on the cotyledons. Microscopical examination reveals a white peridium with an erect margin. The outer wall is 3-8μ thick and the inner wall 5-8μ thick. The peridial cells are rhomboidal in shape, measuring 20-28 x 25-40μ. The aeciospores are spherical or rounded polyhedral, occasionally elliptical, measuring 15-27 x 20-32 μ. They have a closely verucose, colorless wall, measuring 1-2μ in thickness. Fine diagonal striations may be seen in this wall. The contents of the cell are bright orange yellow with many large vacuoles.

<u>Uredinia</u>-

The uredinia are mostly hypophyllous and are borne in scattered round pustules 0.1 to 1.0 mm. in diameter, naked early in their development with the ruptured epidermis plainly evident. They are dark reddish brown in color and appear dusty to the naked eye. The urediniospores are flattened to globoid, measuring 20-27 x 25-30μ. The wall is 1-2μ thick, of a dark, cinnamon brown color and more or less echinulate. It contains 2 equatorial pores.

Telia-

The telia are chiefly hypophyllous. They may be scattered but are usually crowded into irregular groups 2-10 mm. across and occasionally from solid sheets over 2.5 square centimeters of leaf surface. On the petioles and stems they are crowded into long, narrow groups 0.5 - 1.0 x 3.0 - 12.0 mm. When scattered on the leaf surface the telia are 0.4 - 1.5 mm. in diameter. In contrast to the powdery uredinia we find the telia very compact. The pustule appears chocolate brown to nearly black, with an inconspicuous ruptured epidermis. The compound teliospore is made up of two cells which are ellipsoid or oblong. The teliospores measured 20-36 x 36-56 μ in 600 measurements, agreeing almost exactly with Arthur (5). I found the modes of width and length to be 26 x 44μ. They are borne on a colorless pedicel from 70 to 150 μ in length, which remains attached to the spore. The compound spore is slightly constricted where the two cells meet. The upper cell is more obtuse or rounded, while the lower cell is somewhat narrowed. Woronin (33) found that each single cell has a thick outer wall or exosporium of a cinnamon brown color which consists of two layers. The inner layer, he said, usually appears darker and is of uniform thickness while the outer layer stands out plainly at the forward end and has thickenings especially in the extra thickened forms at the apex of the upper cell and at the side immediately below the septum.

Though these cells are very closely united and never separate from each other in nature, Woronin separated the cells from each other by the use of concentrated sulfuric acid. Under this treatment the outer exosporium is decolorized, swells up, and may be seen as an unbroken membrane surrounding the whole teliospore.

A germ pore is evident in the exosporium at the strongest thickened part of each cell of the teliospore. Beside the dark colored exosporium, each cell possesses also a thin and entirely colorless membrane noted by Woronin, the endosporium. Woronin also noted the strongly refractive spot which is found in the fine grain plasmotic contents of each cell but he was not sure whether it was an oil drop or a nucleus. By the use of intravital stains I have found this to be the nucleus.

Promycelia and Sporidia-

Normal promycelia are about 10-12 x 70-80μ in size divided into 4 cells, each of which are 9-12 x 10-16μ. Each of these cells develops an arm or prolongation 5-10μ long at the tips of which develop the ellipsoid to spherical sporidia. These thin-walled spores are densely filled with cytoplasm and measure 6-10 x 9-15μ. Further details on this subject will be found under Germination Studies.

Name of the Causal Organism.

The sunflower rust, Puccinia helianthi Schw. was first described by Schweinitz in 1822. In his Synopsis Fungorum Carolinae Superioris of that year we find three descriptions of the disease. The first mentioned (27) is the aecial stage described under the name of Aecidium helianthi mollis (Schwein.) as "aecium oblong, thick, whitish, with peridia crowded, pale, spores oblong. Frequent on the dorsal side of the leaves of Helianthus mollis, spores under the microscope luteo fuscous, vesiculose, when old, transparent, white." The second and third (28) (29) descriptions are of the telial stage under the genus name of "Puccinia or Dicaemonia." Both are apparently descriptions of the one species as we have it today. He describes "Puccinia heliopsodis" (Schwein.) as follows (28): "Puccinia moderately large, irregular, clustered, bordered by epidermis, chestnut brown, spores oval elongate, longly pedicelate, two-celled. Frequent on the dried leaves of Heliopsidis also Vernoniae. Cells of teliospore equal, septum exactly in the middle of the spore." His third description (29) is under the name of "Puccinia helianthi" which he describes as "Puccinia small orbicular, crowded black, spores globose-oval, two-celled, longly pedicellate. On several common Helianthi, spores fusco luteus, white

transparent pedicel." Ten years later (1832) Schweinitz applies the names Caeoma (Aecidium) helianthatum (Schw.) and Caeoma (Aecidium) tracheliifoliatum (Schw.) Peck (22) in 1885 applied the name Puccinia Viguirae while in 1898 Kuntze used the names Dicaeoma helianthi, Dicaeoma heliopsodis, and Dicaeoma viguirae. The name Puccinia helianthi-mollis was given by Jackson in 1918, while Arthur named the fungus Dicaeoma helianthi-mollis (Schw.) Arth. in his work on the Uredinales being published in the North American Flora commenced in 1907 and not yet completed in 1922. Part 6 containing the sunflower rust was published in February 1922.

The synonomy of the fungus is as follows:

Aecidium helianthi-mollis (Schw.)

Synopsis Fungorum Carolinae Superioris 1:68, 1822.

Puccinia heliopsidis (Schw.)

Synopsis Fungorum Carolinae Superioris 1:72, 1822.

Puccinia helianthi (Schw.)

Synopsis Fungorum Carolinae Superioris 1:72, 1822.

Caeoma (Aecidium) Helianthatum (Schw.)

Transactions American Philogenetic Society 2:292, 1832.

Caeoma (Aecidium) tracheliifoliatum (Schw.)

Transactions American Philogenetic Society 2:309, 1832.

Puccinia viguirae (Peck)

Bulletin Torrey Bot. Club 12:35, 1885.

<u>Dicaeoma helianthi</u> (Kuntze)

Revue General 3^3:469, 1898.

<u>Dicaeoma heliopsodis</u> (Kuntze)

Revue General 3^3:469, 1898.

<u>Dicaeoma viguicae</u> (Kuntze)

Revue General 3^3:471, 1898.

<u>Puccinia helianthi mollis</u> (Jackson)

Brooklyn Botanical Garden Memoirs 1:250, 1918.

<u>Puccinia helianthorum</u> (Rav.)

Fg. Cr. Exs. 90.

<u>Dicaeoma helianthi-mollis</u> (Schw.) (Arth.)

North American Flora 7:427-428, 1921.

<u>Puccinia helianthi</u> (Schw.) is the name which is in most general use, was used in one of the three original descriptions of the fungus by Schweinitz and it seems advisable for convenience and clarity to retain this name in this thesis.

Germination Studies.

The teliospores formed in the fall have not been found to germinate at once but apparently require a rest period of two to three months. The first teliospores which I succeeded in germinating without artificial stimulus formed their germ tubes on November 18, 1921. Previous to this I had succeeded in causing them to germinate by treating leaves covered with pustules with

Chlcroform vapor for one minute and then soaking them for five minutes in one tenth of one percent acetic acid (H_2CO_3). These germinated but formed germ tubes only. After November 18th the teliospores germinated readily the remainder of the winter and throughout the spring as long as the tests were made up to the 10th of July. The teliospores will germinate at a temperature of 6°C. and trash exposed to the elements was found to be germinating as soon as the snow was melted away from it. Woronin (34) states that the first succeeded in germinating teliospores in the first days of February and that germination continued rapidly until the middle of May but that the teliospores which had been preserved for a longer time (from fall to the middle of July) began to lose their viability so that those sown July 19th required three days to germinate while earlier in the season they germinated in ten to fifteen hours. Teliospores which had been kept more than a year he found incapable of germination.

Teliospores which I preserved in the laboratory on the leaves of Helianthus annuus germinated over 95% in early May but decreased rapidly in germinating power. Teliospores placed on water in a beaker on July 6 and kept cool in running tap water showed no germination for the first four days and only two were found which had formed promycelia on July 12th out of over 400 spores which were examined.

Most of my studies on promycelia and sporidia were made in late April and through the month of May. Germination commenced in about 18 hours under room temperature. In the first experiments the teliospores were scraped from the leaf pustule into a watch glass and thoroughly mixed with distilled water to separate the spores in the compact pustule from each other. These were then transferred to the surface of hanging drops which were suspended on VanTieghem rings which were then sealed with petrolatum. These teliospores on germinating gave only long germ tubes without regard to their position whether on the surface or immersed in the hanging drops, attaining in some cases five or six times the length of the entire teliospore and apparently only ceasing growth when all the food material stored in the cell was utilized. Germination seemed more often to commence in the upper cell. The experiments were repeated but the dry teliospores were carefully dusted from the pustules onto the surface of hanging drops without being first wet over in the watch glass. The growth continued to be confined to germ tubes even though the dry teliospores were carefully sown on the surface of the hanging drops, as long as the sealed rings were used. Several thicknesses of filter paper were then placed in the bottom of a petri dish with holes cut through them about the size of the rings. The filter paper was then soaked with distilled water and, after sowing the dry teliospore on the hanging drops, the cover glasses were placed over the holes in the filter paper. A few cases were observed here like (d) Plate I, where sporidia were formed. Excessive moisture and lack of air seemed to be the factors encouraging formation of germ tubes rather than promycelia and sporidia. Another factor might have been the large numbers of yeasts which developed in the hanging drops in spite of using sterile glassware and sterile distilled water. Some sterile agar containing no nutrients was next used as the medium on which to germinate the teliospores. The agar was liquefied in a water bath and clean slides were dipped into the agar with a pair of forceps. The agar hardened at once in a thin uniform layer over the slide and while still warm the excess was removed from the edges and from one side of the slide, leaving the one surface untouched. Slides prepared in this way were placed on several layers of moist filter paper in deep culture dishes. The dry teliospores were then dusted over the moist agar to which they adhered. In about 24 hours promycelia and sporidia like those in (h) and (j) developed. Though conditions of moisture and air were nearly ideal the promycelia were still not the form we think of as characteristic of the rusts. The experiment was again repeated with part of the deep culture dishes at room temperature while the others were placed in a running water bath at a temperature of 12-15°Centigrade. The warmer culture developed more rapidly than those in the water bath and were the same form as before while quantities of characteristic promycelia were formed in those at the cooler temperature of the water bath. One of these normal promycelia is pictured as (i) in the accompanying Plate I. In some cases germ tubes had commenced apparently the formation of sporidia on a normal promycelium, but when left under the microscope still under the same moisture and air conditions but with room temperature these normal promycelia reverted to germ tubes. Two camera lucida drawings of the same teliospore at 11:30 A.M. (k) and 3:00 P.M. (l) of Plate I illustrate this point.

Attempts were made to study the nuclear behavior during the development of germ tubes or promycelia by the use of methylene blue. Figures (a) and (e) Plate I, illustrate how the nucleus, stained blue by the intravital stain, passes out of the cell in all cases observed before the reduction division takes place which results in four cells, each of which produces a sporidium. Further studies of the nuclear divisions following the method of Blackman (8) (9) have been unsuccessful up to the present in showing the details of the nuclear behavior but it seems certain that the nucleus passes out into the promycelium before undergoing either the heterotypic or the homotypic division. The sporidia which were observed in the process of germination developed only germ tubes and not secondary sporidia as is often observed in this group. Woronin describes the promycelia as a colorless or with a faint reddish colored protoplasm (34). The writer has never been able to distinguish with certainty this reddish color in the many promycelia observed.

Wintering and Dissemination of the Parasite.

Trash-

It has been noted by European investigators that one of the greatest sources of infection of Puccinia (Dicaeoma) helianthi (Schw.) is the trash from the previous year's crop. In my examination of fields rust has always been found to be more abundant in fields replanted to sunflowers and in fields adjacent to those which bore a crop of sunflowers the previous year. In the summer of 1919 a large plot of the Kaerpher variety of sunflower was grown in the east half of field #9 at the Michigan Agricultural College. The following year (1920) head rows of selected heads being tested by the remmant system were planted just west of the previous year's plat in the same field. The portion of the field used in 1919 had been planted to wheat in the fall of 1919. Over twenty sunflower seedlings bearing the aecial stage on their cotyledons were found in this wheat field. In one case a portion of a sunflower head had evidently been buried, for a dense cluster of seedlings came up with numerous aecial infections on their cotyledons. The first uredinia were found at the end of the 1920 head rows which were nearest the plot of the previous year, and three weeks before other plots of sunflower were marked by more than occasional pustules the head row series were thickly covered with the summer rust pustules.

<u>Wild Sunflowers</u>-

A second method of spread of the rust fungus is by the many wild varieties of sunflower which are found in this country. Alexander divides the wild forms into 500 species, many of which are doubtless hybrid types. Cross inoculation experiments have resulted in infection of all these wild forms with the exception of *Helianthus tuberosus* by the spores from the cultivated *Helianthus annuus*. Inoculation with the rust spores from *Helianthus tuberosus* as well as all the other wild forms available

have resulted in infection upon the Helianthus annuus of cultivation. Woronin ran a number of cross-inoculation experiments (34) with other genera of the Compositae which are infected by forms closely resembling Puccinia helianthi. He tested all these forms in inoculation tests on H. annuus and found Puccinia discoidearum (Schlecht) which Magnus (20) had previously shown to be nearest Puccinia helianthi in structure was capable of forming the typical rust infection upon Helianthus annuus. He collected the teliospores of Puccinia discoidearum (Schlecht) on Tanacetum vulgare and these sown on healthy Helianthus annuus plants infected the latter, forming pycnia and aecia indistinguishable from those of Puccinia helianthi. These aeciospores he showed to be capable of infecting Helianthus annuus, developing a mycelium which bore the typical uredinia of Puccinia helianthi. Woronin thus proved the surmise of P. Magnus (20) that the rust of Helianthus annuus may be caused by Puccinia discoidearum but he notes the growth is not as strong as is obtained by sowing the teliospores of Puccinia helianthi Schw. Woronin (34) therefore names the species of Tanacetum artemisia and Chrysanthemum on which Puccinia discoidearum usually develops as possible agents in disseminating the sunflower rust.

The rust pustules of Puccinia helianthi are not only found on the involucre and involucral bracts of the sunflower head, but are also in some cases found on the flowers themselves, though this infection rarely extends far from the ray flowers of the head. Here we have another possible source of dissemination for it seems not unlikely that the seed might carry upon their testa some of the teliospores while in practically all samples of commercial seed may be found a considerable amount of small particles of the floral parts and the involucral bracts. The presence of pustules on some of this accompanying material was evident with a hand lens. The attempt was then made to centrifuge the seeds themselves after a thorough steeping in water. This proved unsuccessful but upon shaking the seeds first in alcohol and centrifuging the washings there were a few teliospores found. This was evidently due to the gummy material on the testa to which the teliospores adhered until alcohol was used to dissolve the gum. The presence of teliospores was also demonstrated by scraping the seed with a scalpel and mounting the scraped material in water under the microscope. Although this demonstration of the presence of the teliospores on the seed made seed dissemination very probable, it remained to prove the fact that these seed-carried teliospores were capable of causing the spring infection of the sunflower seedlings. For this experiment a coldframe not previously used for sunflowers

was carefully cleaned out and new soil from a field where no sunflowers had grown was brought in and a layer 18 inches deep placed in the coldframe. Certified seed of the Mammoth Russian variety of Helianthus annuus grown in Missouri was purchased from the Michigan State Farm Bureau and used in this experiment. The soil was moistened with a sprinkler and the seed sown thickly over the surface of a bed 4 x 4 feet and raked in. The frame was kept covered with glass, except at the time of the daily watering with sprinkling can and the examination. About twenty days after the seeding a thorough examination revealed the presence of seven plants which bore aecial cups on their cotyledons. Several of these cotyledons were placed in Gilson's fluid, washed, dehydrated, infiltrated, imbedded and sectioned to prove beyond a doubt that they were the aecial cups of Puccinia helianthi. We thus have positive evidence that in addition to trash other species of Helianthus and a few related genera of the composites we have dissemination by seed.

Varietal Resistance.

Up to the very present time very little work has been done on the breeding of a rust resistant variety of sunflower, although there appears to be considerable

variance in the resistance of different varieties and strains. A variety test for rust resistance was commenced by Professor Spragg of this institution in 1918 (30). The four varieties used were the Mautica developed by Luther Burbank, the Kaerpher from South America, the Mammoth Russian from the Vaughn Seed Store, Chicago, Illinois, and a double mixed H. chrysanthemum from W. W. Barnard Company, Chicago, Illinois. He reported the double mixed and the Burbank were nearly killed by the rust in 1918, the double American variety being outstanding in its rust resisting power, and the Mammoth Russian only fair. In 1919 one plant selection of Burbank, and two each of the South American and Russian varieties were planted in duplicate series with the South American as check. The selections from Burbank and Russian were practically killed by rust, the South American being the only variety that stood up under the attack of rust. Plate II shows the Mammoth Russian and South American at late harvest time in 1919. The South American still holds its green leaves while the Mammoth Russian is dead.

This South American variety is not resistant in the true sense of the word, but may be described as tolerant to the rust fungus. Many large pustules are

formed upon the leaves of this variety, but due to its tolerance the leaves remain green, in spite of the heavy attack, for a much longer time than the common Mammoth Russian sunflower. Work by Satsiferov on the inheritance of the sunflower shows that the ordinary field sunflower of Russia is very much mixed genetically, and it should be possible from this heterozygous mixture to select more resistant strains of sunflower. Satsiferov (26) states that in his studies in 1912 and 1913 all samples of sunflower were attacked gy rust with the exception of the silver-leaved sunflower (Helianthus argyrophyllus Alef.) in samples of which in 1912 not a pustule of the urediniospores or teliospores could be found. The German summary states that this species was found to be entirely resistant but in inoculation experiments I found it possible to infect Helianthus argyrophyllus with teliospores from both the domestic and from wild sunflowers. I found, on having the original Russian article translated (26) that though resistant in 1912, in the wet summer of 1913 rust was very prevalent and that on nearly all samples of silver-leaved sunflower collected in the fall, there were rust pustules. Satsiferov says "The resistance is evidently due to the hairy covering which prevents the spores from entering through the stomata. This gives hopes of developing a more resistant form having the hairy coat of the silver leaf together with the desirable characters of the field sunflower."

Satsiferov attempted such crosses in 1913 but found the late blossoming maternal plant (the silver-leaf) required the use of a late blossoming field sunflower. His crosses failed to mature seed on this account and so were unsuccessful. He mentions the Jerusalem Artichoke (Helianthus tuberosus) as being perfectly resistant to rust in Russia, but this tuber forming sunflower does not bloom in Russia, making that cross impossible. In this country we have the sunflower rust or a strain of it infecting tuberosus heavily.

During the summer of 1919 I had a test plot containing 19 strains of sunflower, one of them being head selections of the Kaerpher South American variety from Professor Spragg's plat of the previous year selected for superior disease resistance. The other species, Helianthus cucumerifolius, Helianthus globosus, Helianthus fistulosus, Helianthus nanus, the double mixed tested by Spragg were all previously tested in the greenhouse by inoculation and found to be all more or less susceptible to the urediniospores from both the Helianthus annuus and Helianthus tuberosus. As the accompanying table shows, none of these varieties exhibited rust resistance though the wild forms were more resistant than the cultivated Helianthus annuus. The most heavily rusted variety

Diagram of Experimental Plot.

<table>
<tr><td colspan="2">3</td><td colspan="2">2</td><td colspan="2">1</td></tr>
<tr><td colspan="2">6</td><td colspan="2">5</td><td colspan="2">4</td></tr>
<tr><td>10</td><td>9</td><td colspan="2">8</td><td colspan="2">7</td></tr>
<tr><td>16</td><td>15</td><td>14</td><td>13</td><td>12</td><td>11</td></tr>
<tr><td colspan="4">19</td><td>18</td><td>17</td></tr>
</table>

Table I -- Tests of Sunflower Varieties.

No.	Species.	Variety or Strain.	Germ. %	Infection.	Ht.	Stand. %	Oct. 10 Maturity.
1.	H. annuus	Kaerpher (selected heads.)	80	Light	3'	80	Fair
2.	"	" "	50	Mod.	3'	50	"
3.	"	" "	85	Traces	2'	85	"
4.	"	" "	70	Mod. heavy.	4'	70	"
5.	"	" "	60	Mod.	5'	60	Good
6.	"	" "	60	Mod. heavy.	5'	60	"
7.	"	" "	60	"	7'	60	Fair
8.	"	" "	70	"	7'	70	"
9.	"	" "	85	Light	5 1/2'	70	"
10.	"	" "	2	Traces	2'	2	
11.	"	Burbank's Prolific White (Mantica)	90	Mod. heavy.	2'	60	1st-Imm.
			90	Heavy.	5 1/2'	70	2nd-Fair
12.	H. cucumerifolius	Grandiflorus Stella (Dreer)	20	Light.	1'	20	1 Imm.
13.	"	"	30	Trace	1'	30	Good
14.	H. globosus fistulosus	"	75	Light	4'	75	Fair
15.	H. nanus Fl. Pl.	"	85	Mod.	1 1/2'	70	"
16.	Helianthus sp.	Trade name (Japanese)	20	--	--	--	Good.
17.	H. chrysanthemum Dbl. Mixed	Spragg Acc. 4	0	--	--	--	--
18.	Helianthus sp.	Trade name (Texas-Winged Taubenhaus)	0	--	--	0	--
19.	H. annuus,	Mammoth Russian (F.Bureau)	90	Mod. heavy.	5'	90	Row good
			90	"	9'	90	Patch "

#12 and 14 many heads.
#19 25% of plants many heads.

was the Burbank, both in the field and in the greenhouse, and on account of its great susceptibility to the rust, at least in this locality, the Burbank variety was used in the greenhouse for all inoculations in studying the morphology of the rust fungus.

Control.

Since one of the greatest agencies in the dissemination of Puccinia helianthi is the trash of leaves and stems from the previous year's crop, sanitation at once presents itself as one of the most important means of control. Where sunflowers are used for silage the entire plant is ensiloed, this ridding the field of practically all the trash. Where grown for seed the trash should be removed and burned, and in no case should the same field be used for sunflowers two years in succession. Woronin (34) found the teliospores are not viable for over one year and my results confirm this statement, so that a rotation which permits one year or more to elapse between successive plantings of sunflower should control this source of infection. Sporidia are known to carry long distances so that it is advisable not to grow sunflowers in a field adjacent to a field which bore a crop of sunflowers the previous year. (See my results under dissemination.)

To deal with the dissemination by other related species of plants, the wild forms of sunflower and also species of Tanacetum, Artemisia and Chrysanthemum should be destroyed.

Woronin (33) suggested the burning of the previous year's rubbish and the avoiding of successive seedings, in 1872. He also advocated a careful plucking out of the sunflower plants showing the orange rust or spring stage and the planting of sunflowers in rows instead of broadcast to slow up the spread of the rust. This latter is not practical since the labor and expense would be enormous.

Karsin in a popular type of bulletin in the Russian language, in 1896, repeated these suggestions and, among other ideas, brought forward the idea that soaking the seed in lime water or blue vitriol might kill the spores of the rust and aid in the control of the disease.

The writer conducted germination experiments and found 1/20 of 1% copper sulphate would prevent the germination of the teliospores. The effect of copper sulphate solution on the viability of the seed was then tested in numerous experiments. Solutions of copper sulphate varying in strength from 1/2 of 1% to 10% were used and the time of soaking of the seed varied from

1/2 to 3 hours. The germination was only impaired in cases of excessively long soaking when the seed commenced to germinate in the copper sulphate solution. When this occurred, the radicle blackened and the seedling died, but in all cases of under six hours soaking there was no effect upon the viability of the seed. In fact, the germination was faster and reached 100%, while the untreated check seed was about 95%, due to the beneficial action from the presoaking.

For all purposes, a 1% solution seems advisable with a 30-minute period of soaking, but no injury is done if the seeds are left in the solution for several hours. The most convenient method for treating the seed is as follows:

Weigh out the copper sulphate and place in a small cloth sack. Heat a small quantity of water and stir with the sack of copper sulphate, or suspend near the surface of the hot water. In a few minutes the copper sulphate will be dissolved. A ten gallon stone crock is an excellent receptacle. Pour the water into the crock and add the copper sulphate solution. Pour the seed into the solution and press them under the surface of the solution with a cover cut to fit the crock. Weight the cover to keep the seed immersed and leave for 30 minutes. Then remove the seed and spread in the shade to dry. A bushel may be treated at a time in this way.

Treated seed was planted in a plot adjoining untreated seed at Chatham Experiment Station in the Upper Peninsula of Michigan. The treated plot at Marquette, Michigan did not develop rust until 20 days after the untreated plot, but by fall the uredinio-spores had spread to such an extent from the untreated plot that there was little difference between them. One of the County Agents, Mr. C. E. Atwater of Gladwin County, also reported that the rust developed much later and to a less degree on the plot of treated seed which were sent to him from this station. He considered the seed treatment a great success and wished to treat all his seed this year.

Some treated seed was planted with corn in Lenawee County by a farmer for silage, and at time of harvest the sunflower plants were remarkably free from rust, with even the lower leaves large and green, with but an occasional pustule of rust.

Sunflower seed treated with a dust treatment instead of soaking was planted in the silage corn at the Michigan Agricultural College farm and the plants on July 10th showed no rust whatever, while the untreated plants in Professor Spragg's breeding experiments were quite heavily rusted although they were of the Kaerpher variety. The experiment is being repeated this summer with copper sulphate dipped seed, seed dusted with Mackie dust, and seed dusted with Corona dust by a number of County Agents and also by a group of farmers in Lenawee County. Mackie dust consists of 50% dehydrated $CuSO_4$ and 50% hydrated lime, while Corona dust contains about 18% $CuCO_3$ and a large percentage of $CuSO_4$.

It seems likely that we may find a control in the form of a rust resistant variety by systematic plant breeding. Professor Spragg's rust-resistant sunflower is in reality a rust tolerant race of sunflowers from which it may be possible to separate a superior variety.

At the present time the best control seems to be a proper rotation of crops together with seed treatment, and while this will not entirely control the rust, it reduces the severity of the attack to a small amount and will pay in the increased tonnage and succulence of the silage and in the larger production of seed which give sunflower silage its high feeding value.

Conclusions.

Puccinia helianthi is an autoecious rust with five spore forms, but which occasionally elides the aecial stage. The rust infects all species of Helianthus and is economically important on the cultivated varieties of Helianthus annuus. The organism is carried over from year to year by teliospores on the trash of the previous year's crop and upon the seed. The attacks of the fungus can be, for the most part, controlled by rotation and by seed treatment. The best treatment, so far as experiments have gone, consists in soaking the seed for thirty minutes in 1% copper sulphate solution. No true resistant varieties of the cultivated Helianthus annuus have been developed up to the present time.

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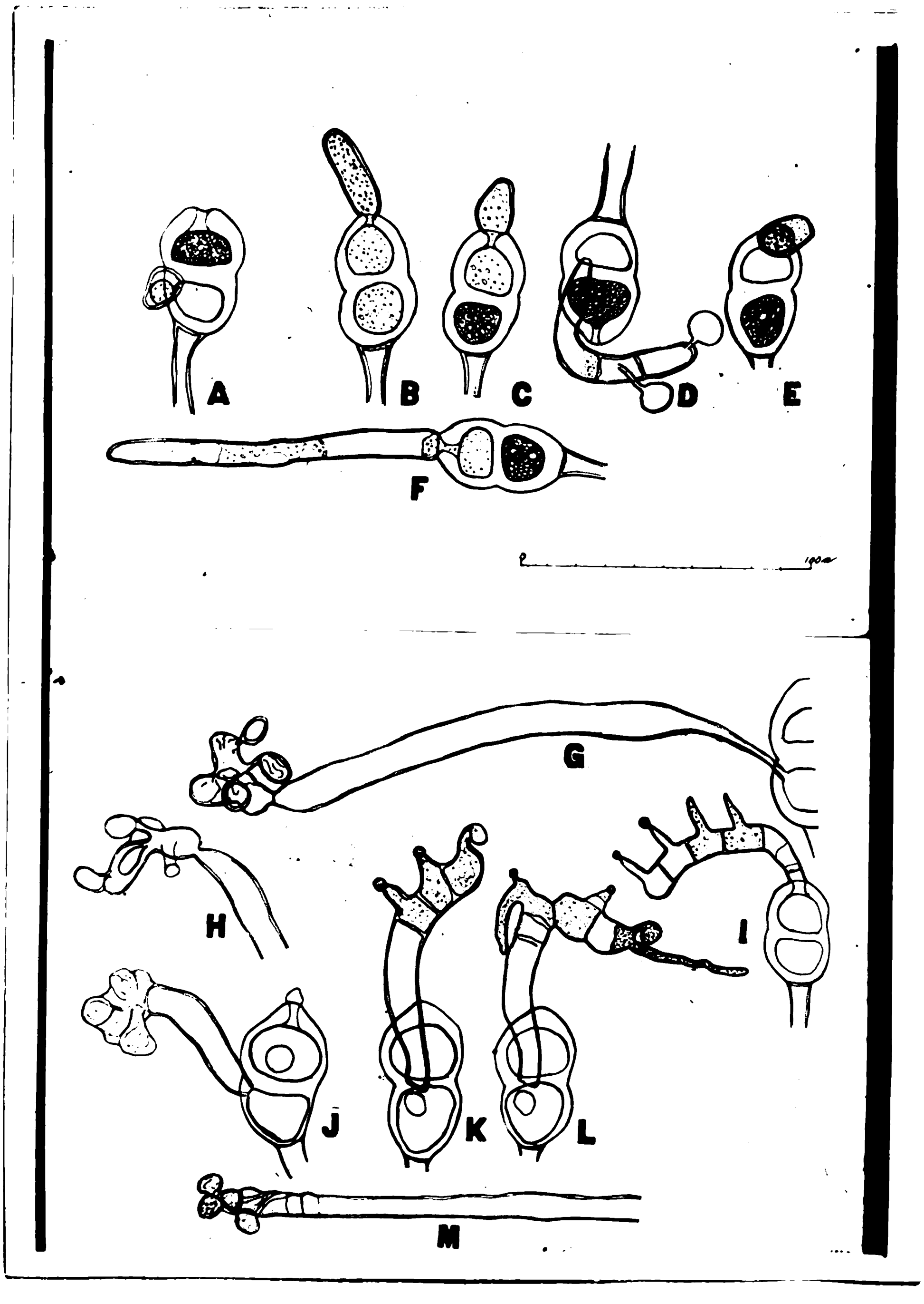


Plate I. Camera lucida drawings of germinating teliospores.

A-Germ tube showing nucleus. B & C-Early stage of development of germ tubes. D-Promycelium bearing two sporidia. E-Germ tube showing nucleus. F-Teliospore showing germ tube which did not form sporidia. G-Germ tube growing out of drop of water, sporidia formed at end. H-Promycelia developed at room temperature on moist agar. I-Normal promycelium developed at 14°C. on moist agar. J-Promycelium developed at room temperature on moist agar. K-Promycelium developed at 12-15°C. on moist agar. Drawn at 11:3- A.M. L-Same promycelium at 3:00 P.M. after 3 1/2 hrs. at room temperature. M-Germ tube with end protruding from a drop of water forming terminal sporidia.

Plate II. Plat showing Mammoth Russian and South American sunflowers in trial plat of Professor F. A. Spragg. The South American, at the right, retains its leaves, while the Mammoth Russian, at the left, is practically killed by the rust.

Plate III. Pycnia on leaf of Helianthus annuus.

Plate IV. Aecial cups on leaf of Helianthus annuus.

Plate V. Aecial cups on leaf of Helianthus annuus.

Plate VI. Aecial cups on leaf of Helianthus annuus.

Plate VII. Aecial cups on leaf of Helianthus annuus.

Plate VIII. Leaf of Helianthus annuus severely infected with the telial stage of the rust.

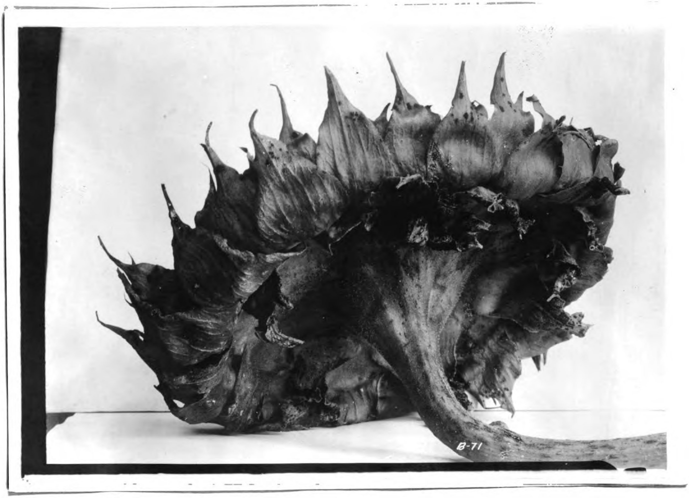


Plate IX. Head of Helianthus annuus showing the pustules of teliospores on the involucre and involucral bracts.

Plate X. Frequency curves of width and length of teliospores of <u>Puccinia helianthi</u>.

Plate XI. Frequency curve showing size of teliospore of <u>Puccinia helianthi</u> and <u>Puccinia discoidearum</u> (Mag.)

Plate X.

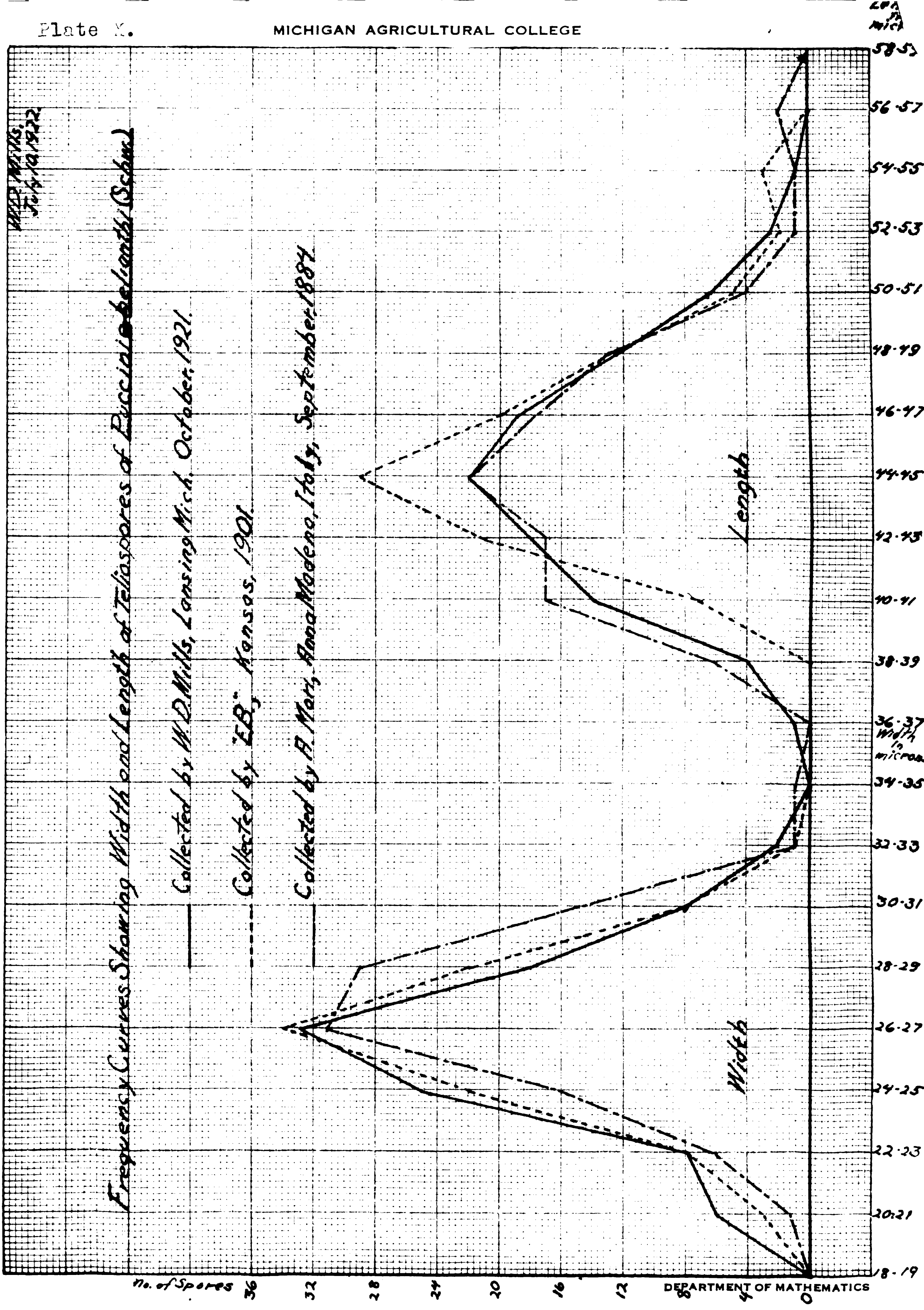
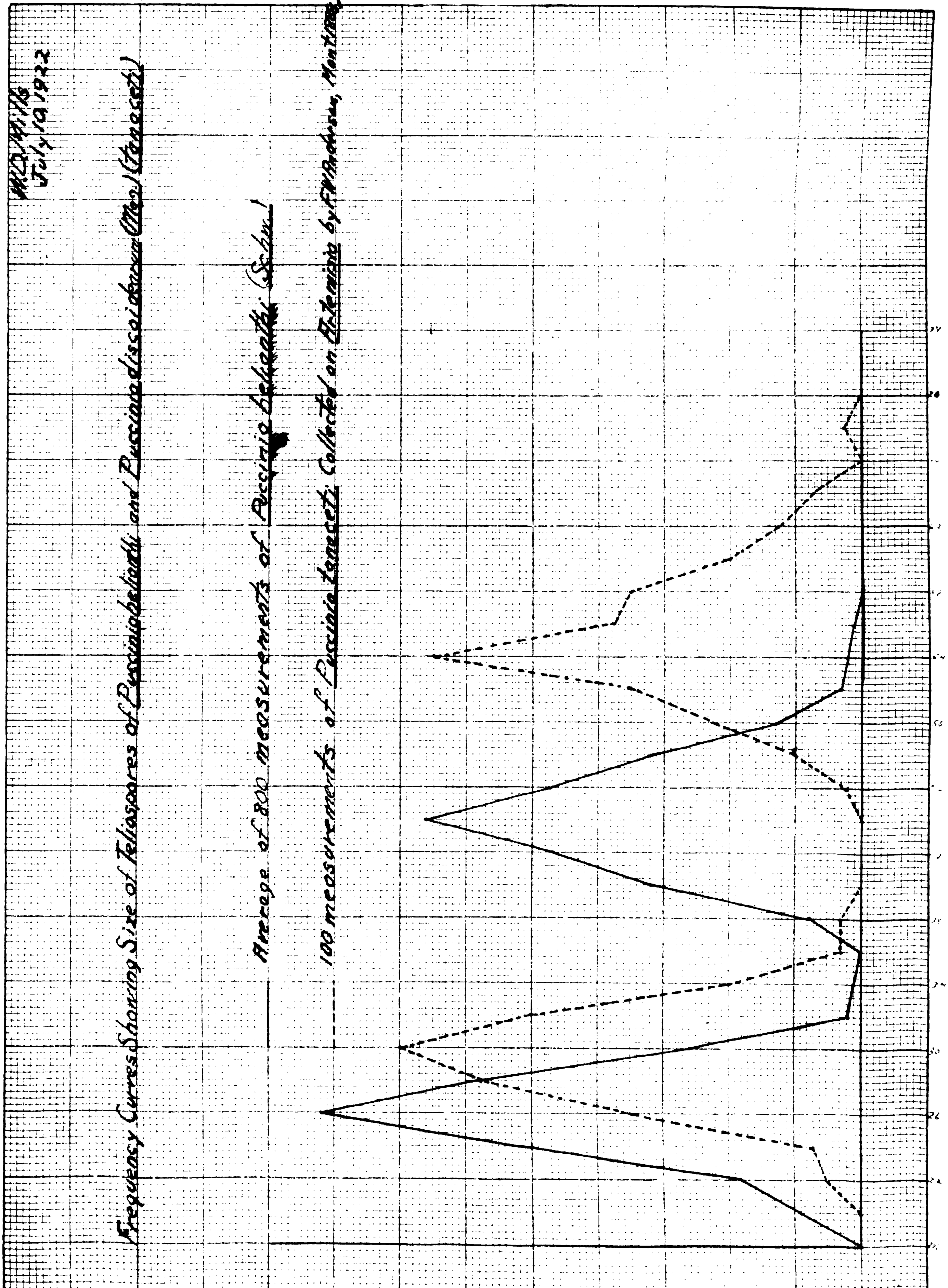

W.D. Mills,
July 10 1922
Frequency Curves Showing Width and Length of Teliospores of Puccinia helianthi (Schw.)
Collected by W.D. Mills, Lansing Mich. October 1921.
Collected by "E.B." Kansas, 1901.
Collected by A. Mori, Anna Modena, Italy, September 1884
Length
Width
No. of Spores
36
32
28
24
20
16
12
8
4
0
18-19
20-21
22-23
24-25
26-27
28-29
30-31
32-33
34-35
36-37
Width in microns
38-39
40-41
42-43
44-45
46-47
48-49
50-51
52-53
54-55
56-57
58-59

Plate XI.



W.D. Mills
July 10 1922
Frequency Curves Showing Size of Teliospores of Puccinia helianthi and Puccinia discoidearum (Mor.) (Tanaceti)
Average of 800 measurements of Puccinia helianthi (Schw.)
100 measurements of Puccinia tanaceti Collected on Artemisia by F.W. Anderson, Mont. 1888