A NEW LEAF SPOT OF VIOLA CUCULLATA.

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A leaf spot on *Viola cucullata* has been prevalent in Indiana and neighboring States for a number of years. It is especially noticeable during the early spring months. Collections of leaf spots on this host in different parts of the country have been made from time to time and have been filed away in the herbarium without being classified or wrongly labeled as the Phyllosticta leaf spot. A careful examination during the past year has revealed the fact that this particular leaf spot is caused by a Colletotrichum which has never been described as occurring on this species. Since this disease is widespread it was thought worth while to make a careful study of the causative organism.

Of the violets which occur in this region only *Viola cucullata* has been found to be attacked by this particular fungus. It is interesting to note that while this species is attacked *V. palmata* is apparently immune. *V. cucullata* was formerly considered a variety of *V. palmata* and the immunity of the latter emphasizes the specific difference. However, only a limited number of plants of *V. palmata* have been observed and these in a region where the disease was not common on the other species.

Cultivated violets have been examined only in the local greenhouses. It is probable, however, that all cultivated species are immune, otherwise the disease would have been observed and reported by those especially interested in violet diseases.

Macroscopical Appearance.—The fungus produces a typical leaf spot. The earliest indication of infection is a pale area with a definite dark green border. Later the area in the center of the spot dies, turns white, grey or light brown, a dark brown ring appears about the edge, forming a definite, regular spot. At an early stage the acervuli appear as dark brown dots on the lighter central area. They are irregularly arranged and occur on both sides of the leaf. The dark color of the acervuli is due, in part, to the numerous setse. Later the center of the spot becomes very thin and papery and may fall out, thus giving a shot hole effect. When badly infected the spots are occasionally confluent. Usually there are only a few spots on a leaf.

Etiology.—The fungus gives rise to numerous acervuli, which are dark brown or black, irregularly scattered, varying greatly in size (50-200 microns). They are beset with dark brown seta. The setae are numerous, arising from any part of the acervulus, 1-4 septate, dark brown, sharppointed, straight or slightly curved above the base. The base is usually bent or curved in various ways.

The spores are hyaline, non-septate, slightly curved averaging 4.5x25 microns. They are borne on short, hyaline conidiophores. In some cases at germination a delicate septum was observed in the middle of the spore. This is by no means always present.

Nomenclature.—There has been, in the past, some confusion in regard to the limits of the genera Collectorichum, Vermicularia, Volutella and Chaetostroma. This has been due, largely, to the lack of care exercised by investigators when species of these genera have been studied. Sections carefully made clear up generic confusion very easily. The fungus described above is undoubtedly a Collectorichum since there is no pycnidium, the spores being borne on short conidiophores in a setose acervulus.

In 1899, Dr. Ralph Smith' described a leaf spot of pansy caused by a Colletotrichum. The type material of this fungus has been examined and found to differ from the Colletotrichum under discussion in the size and shape of the spores, the shape of the setæ and the character of the spot produced.

Dr. Peck² in 1878 described a leaf spot of *Viola rotundifolia* as follows:

"Vermicularia concentrica Peck and Clinton n. sp. Perithecia small, black, beset with straight, rigid bristles, concentrically placed on arid, orbicular spots; spores oblong, slightly curved, pointed at each end, colorless, .0008'-.001' long.

"Living leaves of Trillium crythrocarpum. Pine Valley, Clinton, July:

"The tissues at length fall out from the affected spot, leaving apertures through the leaf. The perithecia are less regularly disposed near the center of the spots. Judge Clinton also sends a variety on the leaves of *Violu rotundifolia* in which the concentric arrangement of the perithecia is not at all preceptible, but I detect no other difference."

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⁽¹⁾ Botanical Gazette. 27: 203-204, Mar. 1899.

⁽²⁾ Report of the N. Y. State Botanist 1878. 29th Annual Report of the N. Y. State Museum of Natural History, pps. 47-48.

In his "Sylloge". Saccardo³ changed the specific name of the fungus to *Peckii*. His nomenclature is as follows: *"Vermicularia Peckii* Sace.

F. Concentrica Peek, 29th Ann. Rep. N. Y. State Mus. Nat. Hist. 47-48, 1878. (Not Lev., Ann. Soc. Nat. 66, 1845.)

"V. Peckii var. Violæ rotundifoliæ Sace."

Through the kindness of Dr. H. D. House, State Botanist of New York, 1 was able to secure authentic specimens, collected and determined by Dr. Peck subsequent to his description of the fungus. These specimens were from Trillium crythrocarpum Michx. (T. undulatum Willd.) and from Viola rotundifolia. A careful examination of these specimens, together with some recently collected material from Dr. House, was made. Some of the spots were embedded in paraffin, sectioned and stained. From these examinations it was concluded that the fungus occurring on Viola rotundifolia was not a Vermicularia but was identical with the Colletotrichum occurring on V. cucullata. The spore measurements and general characters of the acervulus, setæ and conidiophores of the fungus on Trillium were also identical but, as stated by Dr. Peck, the acervuli of the former occur in definite concentric circles in the spot, while in the latter no such arrangement is noticeable. Whether or not the species on Viola cucultata is identical with the one on Trillium can only be determined by crossinoculation. Up to the present time the author has not had an opportunity to complete his investigations along this line. It would be unusual. however, to have a fungus of this type parasitic on hosts so widely separated as Trillium and Viola. The identity of the fungi on the two violet species can hardly be questioned. The nomenclature of the fungus on Viola rotundifolia is so awkward and incorrect that a change should be made. However, this is not advisable until the relationship to the fungus occurring on Trillium is definitely settled.

Life History of the Fungus.—'The field observations of this fungus have been limited to a single year. The disease appears at a very early period in the spring on leaves that have evidently lived over winter. The earliest collections were made in the first week in April before the plants had time to develop leaves. It is probable therefore that the fungus lives over winter on the old leaves of the plant, although it has not been observed during the winter months.

^(*) Saccardo, P., Sylloge Fungorum. 3: 232. 1884.

The spread of the disease in the early spring is evident from an observation of an infected plant. A single plant often has at first only one or two infected spots. As the new leaves develop they are seen to be free from the disease, but a few days after a damp period these leaves are badly infected. This disease is, in many respects, an early spring one, for the most luxurious growth of this species of violet is during April and May and the fungus seems to thrive best during cold, damp weather. At no time do the number of spots become so great as to kill the leaf. The damage done by this fungus is evidently negligible so far as this species is concerned.

The fungus spores are probably distributed through the usual agents. They are produced in great numbers on both sides of the leaf and they germinate readily in tap or rain water.

Artificial Inoculations.—A detailed report of the experiments on inoculations made would be out of place at this time. Summarizing these, it was found that infection by sprayed spores on the leaves was readily brought about if the plants were kept very moist for several days. The number of infections were small on each plant. Cross-inoculations on V, public plants were results.

Cultural Characters.—The fungus is easily isolated by poured plates of the spores. Cultures were made on various media, but a description of the development on dextrose-potato agar will be sufficient to show the general character of the growth. The mycelium is at first hyaline, mostly confined to a thick growth on the surface of the agar, with scanty aerial mycelium. At the edge of the advancing mycelium the agar takes on a pink and then a red color which is very striking. Later the mycelium darkens and in some cases within four days conidia are to be found. The darkening of the mycelium continues and within two weeks a heavy, black, stroma-like crust is formed over the surface of the agar. On this stroma hyaline, gelatinous areas of conidia are developed.

Conclusion.—This preliminary report on a leaf spot of *Viola cucullata* is given in order to stimulate others to examine the plants of this species during the coming spring, thus establishing more definitely its range and determining whether or not it is confined to this one host. An effort will be made by the writer to determine the relationship of this species to the one occurring on Trillium and on *V. rotundifolia*.