Green Leaf Spot on Tea Leaf Caused by a *Cercospora*

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It seems that green leaf spot has been distributed in tea plantations in various parts of Japan since ancient days. But in the past the lesion of the symptom was regarded as a rust mite bite or a physiological symptom, and its cause has been investigated in detail and no report has been made.

Hirokawa³⁾ found that this symptom widely attacked tea plantations (Yabukita variety) at Kanaya Machi, Shizuoka Prefecture, in autumn of 1968, causing heavy damage by defoliation.

He felt the necessity of inquiring into the cause of the symptom and considered a counterplan by main studying the pathogenicity of isolated fungi and their taxonomy until 1970. He named as green leaf spot. His studies on the disease are described in the following.

Symptom and attack of green leaf spot

Green leaf spot attacks tea leaves. In the beginning, irregular-shaped small dark green protuberances appear separately or in groups of several to dozens on the back of grown leaves. They are at first about the size of a needle head and enlarge irregularly to a nevus shape as they are protuberant. The lesional part is 1.2–2.1 times as thick as a normal leaf. This increase in thickness is mainly due to cell enlargement in the spongy tissue.

The symptom is estimated to be spread throughout the country. A slight attack of

the green leaf spot is out of the question, but a heavy attack occasionally induced under the influence of weather or nutritional conditions of tea plants does a great deal of damage to tea plantations, accompanying severe defoliation.

Especially, young or ill-fertilized weak tea plantations are apt to be attacked by the disease. A considerable part of fallen leaves at tea plantations in winter are suspected to be attributed to this disease.

Isolation and inoculation of pathogen

A species of fungi belonging to *Cercospora* was isolated from diseased leaves by ordinary techniques as a pathogen of the green leaf spot. In September, the culture of this fungus was put into a mortar and ground with a small amount of sterile water to make a syrupy suspension, and it was applied with a brush to new leaves on two groups of Yabukita tea plants which were grown in different places and different from each other in nutritive condition.

The inoculation was made mainly onto the back of leaves, and each of the treated plants was covered with a polyethylene bag to keep in a moist chamber for two days. Table 1 shows that green spots appeared on all the leaves thus treated.

The same *Cercospora* fungus as inoculated species was easily isolated from these lesions. The incubation period was estimated to be about three weeks. Similar inoculation

Tea plant	Group	Date of inoculation	Date of examination	Number of inoculated leaves	Number of	Degree of infection			
					infected		+	#	##
Variety Yabukita (weak growth)	Inoculated	Sept. 4	Oct. 4	25	22	3	21	1	
			Oct. 22		25		2	22	1
	Control		Oct. 4	5	0	5			
			Oct. 22		0	5			
Variety Yabukita (vigorous growth)	Inoculated	Sept. 4	Oct. 4	14	14		4	10	
			Oct. 22		14		1	11	2
	Control		Oct. 4	6	0	6			
			Oct. 22		0	6			

Table 1. Pathogenicity of the Cercospora sp. on Tea Leaves

Notes: - non-infected, + slight, + moderate, # severe

experiments were also carried out in May and June, showing almost the same results as mentioned above. This symptom is remarkably different from other diseases of tea plants in the point that it can affect old leaves even if inoculated without injuring them.

Morphology of pathogenic fungus

Spores of the pathogenic fungus could not be found on diseased leaves, but the mycelium was estimated to be present. The fungi belonging to *Cercospora* are usually classified by their morphological features observed on host plants, but no organ which could be taken as the basis of classification was found on the green-spotted leaves as mentioned above.

Therefore, eight lines of *Cercospora* (?) fungi isolated from tea leaves (attacked by the green leaf spot and the brown round spot) and camellia were cultured to compare their morphological features with each other in the cultural condition for the purpose of finding a clue to their identification.

These cultured fungi all formed rod-shaped conidia which were characteristic of *Cercospora* and were pointed or a little thickened at the end. The conidia of the fungus isolated from the green spot were $60-80 \times 2.5$ -3.3μ in size and had five-nine septa.

Similar disease of camellia

In 1969, diseased camellia leaves closely resembling the green leaf spot of tea in symtom were found in the grounds of the National Research Institute of Tea at Kanaya-Machi, Shizuoka Prefecture, and a *Cercospora* fungus which formed colonies similar to those of the fungus of tea green spot origin was isolated from them in a high frequency.

The two lines of fungi were different from each other in the form of conidia in the cultural condition, but the cross infection between camellia and tea with these fungi proved successful in contrast to the reports of Johnson et al.⁴⁾ and Berger et al.¹⁾ on the *Cercospora* diseases of the tobacco and forage crops, respectively.

They found the presence of mycelia which were morphologically the same but different from each other in pathogenicity. All these facts seem to suggest that parasitism must be taken into consideration for the classification of *Cercospora* fungi as pointed out by Katsuki.⁶⁾

Defoliation caused by green leaf spot

It is often discovered that tea leaves are apt to be shed when attacked by the green leaf spot. To investigate defoliation caused

Test plant	Group	Date of inocula- tion	Date of exami- nation	Total number of leaves examined	Leaves inoculated after development			Leaves developed after inoculation		
					Total	Number of fallen leaves	Percent- age of fallen leaves	Total	Number of fallen leaves	Percent- age of fallen leaves
Variety Yabukita (weak growth)	Inocu- lated	Sept. 4	Oct. 22	201	102	22	21.6%	99	7	7.1%
			Dec. 1	189	89	49	55.1	100	5	5.0
	Control		Oct. 22	32	18	1	5.6	14	0	0
			Dec. 1	34	17	1	5.9	17	0	0
Variety Yabukita (vigorous growth)	Inocu- lated	Sept. 4	Oct. 22	119	63	1	1.6	56	0	0
			Dec. 1	101	50	2	4.0	51	1	2.0
	Control		Oct. 22	48	26	0	0	22	0	0
			Dec. 1	51	26	1	3.8	25	0	0

Table 2. Defoliation of tea plants by the inoculation of Cercospora sp.

by the disease, an inoculation experiment was conducted in autumn, 1969, by smearing ground mycelia with a brush on three to four leaves from the top of each branch.

The inoculated leaves were labeled to distinguish from new leaves to develop after the treatment. The defoliation of the inoculated leaves was remarkably different from that of the control (uninoculated) leaves or new leaves developed after the treatment as shown in Table 2, and markedly variable according to the vigor in the growth of plants. It was severe in poor growers.

Almost all the inoculated leaves, however, were shed by next March irrespective of the growth of plants. Though such defoliation in inoculation experiments seems to be due to the damage of leaves by an extremely large number of lesions, it is estimated that a great deal of damage is done by defoliation to tea plants even in natural fields conditions when the leaves are heavily infected.

Relationship between green leaf spot and brown round spot in tea

Two diseases caused by *Cercospora* fungi, red round spot and brown round spot, have been described in the tea,²⁾ but the distinction between them is rather obscure in many points.

The symptom of a disease which we habitually call brown round spot is the appearance of brown irregularly round spot on leaves accompanied with the growth of gray to dark green molds (*Cercospora*) on the spots.

Such a symptom, however, often appears together with green spots on the same tea plant or even on the same leaf, and it seems possible that one and the same pathogen causes the different symptoms according to the condition of disease development.

Cercospora fungi are usually classified on the basis of their morphological features compared on host plants, but it is impossible at present to apply this method to the pathogenic fungus of the green leaf spot because no fungous organ useful for the classification can be found on diseased leaves as mentioned above.

Accordingly, it is very difficult to prove the difference between the pathogenic fungi of the two diseases, green leaf spot and brown round spot, by a morphological comparison, except observations in cultural conditions.

When the two pathogenic fungi are morphologically compared with each other on the basis of observations in the cultural conditions and on host plants as well as descriptions published, their characteristics are as follows: The basidia formed on the red round spots cluster on the stroma and have no septum, being 20-40 μ in length. The rod-shaped conidia formed on the basidium are a little curved and 63-118 \times 2.3-3.0 μ in size 1 having 6-11 septa. This observation agrees with the descriptions of *C. theae* by Hara²⁾ and Katsuki³⁾.

On the other hand, the conidia of the fungus isolated from green spots of tea leaves are $6-80 \times 2.5-3.3 \mu$ in size and have five to nine septa, showing close resemblance to those observed on the brown round spots.

From these observations it seems to be quite possible that the fungus found on the brown round spots, that is, *C. theae* is the same species as that isolated by Hirokawa from the green spots of tea leaves.

To examine differences in pathogenicity and disease development among the different lines of *Cercospora* fungi, inoculation experiments were carried out three times under different conditions concerning the inoculation time and the age and injury of leaves to be inoculated.

Cultured fungi derived from the green spot and the brown round spot of tea and the green spot-like lesion of camellia as well as conidia obtained from the brown round spot of tea were used as inocula in these experiments, but in every case green spots appeared on the inoculated leaves irrespective of the difference of inocula.

From this result it seems that the green leaf spot and the brown round spot are different symptoms manifested by the same pathogenic fungus.

Disease-control experiment

In autumn of 1970, field experiments were conducted to test the effects of Propineb and six other chemicals on the control of the green leaf spot. Among the chemicals tested, the following appeared to be considerably effective for the purpose: Propineb (60) on 1:500 ~800 dilution, Anthraquinone (13)—copper sulfate (42) on 1:500 dilution, Difolatan (80) on 1:1000 dilution, Polycarbamate (75) on 1:600~800 dilution and Bordeaux mixture (4-4 formula).

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