Evaluation and genetic analysis of the resistance to tea gray blight in tea genetic resources in Japan.

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Summary

Tea gray blight caused by *Pestalotiopsis longiseta* Spegazzini is a severe disease in tea fields in Japan. It has been clarified that the resistance of tea plants to the disease is controlled by two independent dominant resistance genes Pl_1 and Pl_2.

In this paper the resistance to tea gray blight was evaluated in the tea germplasm preserved at the National Institute of Vegetable and Tea Science in Makurazaki, Kagoshima Prefecture. The genotype of 432 plants preserved as genetic resources were also analyzed by using by many cross combinations. A wide variation of the resistance of tea plants to tea gray blight was observed both in phenotype and in genotype.

The majority of Assam plants (*C. sinesis* var. *assamica*) showed a high level of resistance and very little variation both in genotype and phenotype. However, Japanese native tea plants which belong to *C. sinensis* var. *sinensis* showed a large genetic diversity in the resistance to the disease. In tea plants of var. *sinensis*, the Japanese native plants included wider genetic diversity than the introduced Chinese plants in the resistance to tea gray blight. Since many of the tea plants derived from foreign countries were highly resistant to the disease and had two Pl_1 genes which confer a high level of resistance in many cases, they are very important as materials for breeding cultivars that are resistant to the disease.

Key words

Tea, Genetic resources, Genetic analysis, *Pestalotopsis longiseta*, Gray blight

Introduction

Tea gray blight caused by *Pestalotiopsis longiseta* is a very severe disease as well as tea anthracnose caused by *Colletotrichum teae-sinensis* in Japan. Since it was shown that 'Yabukita' which is presently the leading tea cultivar in Japan is susceptible to the fungus, it was deemed essential to develop tea cultivars resistant to the disease in Japan.

There are considerable differences in the susceptibility to *P. longiseta* among tea cultivars. A method of detecting resistance to the disease has been developed (Hamaya and Horikawa, 1982) that enables a genetic analysis of the resistance to the disease to be carried out. Genetic analysis revealed that the resistance of tea plants to the disease is controlled by two independent dominant resistance genes Pl_1 and Pl_2. The gene Pl_1 which confers a high level of resistance is genetically epistatic in relation to the Pl_2 gene which confers a moderate level of resistance (Takeda, 1988).

The present report deals both with an evaluation of the resistance to *P. longiseta* and a genetic analysis of the resistance to the disease in tea germplasm preserved at the National Institute of Vegetable and Tea Science, in Makurazaki.

Materials and Methods

The resistance of 2,480 plants to tea gray blight was evaluated after inoculation of the fungus in the field. For the inoculation, a mature leaf on a healthy shoots was wounded by the sharpened tip

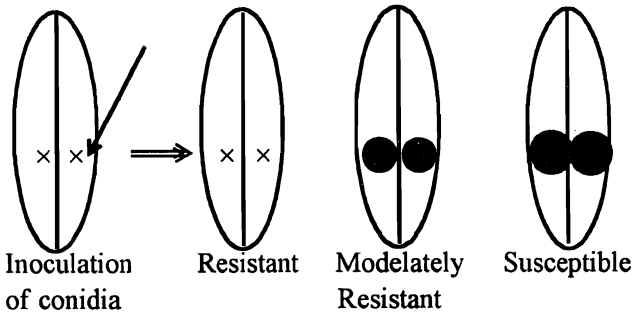

Fig. 1 Lesions of the three groups in the resistance to *Pestalotiopsis longiseta*

of a 3 mm wide (+) screw driver and infected with a water suspension of conidia placed on the tip of the instrument. Since 5 healthy leaves per plant were inoculated in two areas, a total of 10 areas were inoculated. The degree of resistance was evaluated 15-18 days after the inoculation by measuring the diameter of the lesions, and then plants were divided into three groups: resistant (R), moderately resistant (M) and susceptible (S) (Fig. 1).

Moreover, 432 plants and 88 cultivars which belong to var. *assamica*, var. *sinensis* and their hybrids were crossed to the susceptible cultivars as 'Yabukita', 'Saemidori' and 'Asatsuyu' of which the genotype is $pl_1pl_1pl_2pl_2$, and their F_1 progenies were evaluated by the artificial inoculation as mentioned above. Based on the results of the segregation ratio of the F_1 plants according to their resistance to the disease, the genotypes of the parents were analyzed.

Results and Discussion

1. Evaluation of the Resistance to Tea Gray Blight of the Tea Germplasm

Screening tests for the resistance to tea gray blight were carried out for 2,480 plants collected worldwide and maintained in Makurazaki station. The results are shown in Fig. 2.

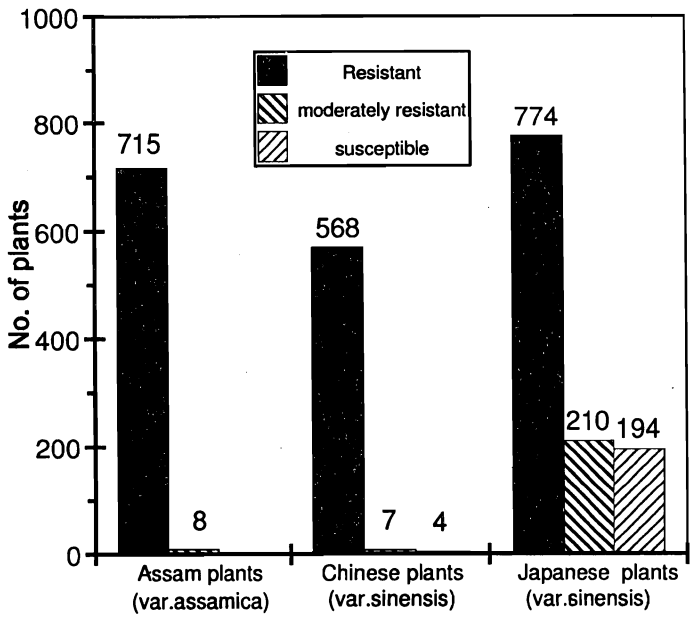

Fig. 2 Phenotypes of the tea plants in the resistance to *P. longiseta*

The Japanese native plants collected from all over Japan showed considerable variation in the resistance. Susceptible plants accounted for 16.5% of the total number of plants which is the same ratio as those having moderate resistance to the disease. There were 774 resistant plants out of 1,178 plants, accounting for 65.7%. In the var. *sinensis* there were large differences between Japanese native plants and introduced Chinese plants which were collected from 'Zhejiang', 'Jiangxi', and 'Anhui' Provinces in China and Darjeeling in India. These foreign materials belonging to var. *sinensis* were generally resistant in phenotype, although some of the materials were moderately resistant and susceptible. In 723 plants belonging to var. *assamica* which were collected from India, Sri Lanka, Myanmar, Vietnam and Bangladesh, all of

the materials showed resistance except for only 8 plants having a moderate level of resistance. There were no susceptible plants in the Assam variety.

2. Analysis of the Genotype of the Tea plants in Resistance to Tea Gray Blight

The genotypes of the resistance to *P. longiseta* were analyzed in 432 plants by the segregation ratio of the F_1 progenies between the susceptible cultivar 'Yabukita' and tested plants (Fig. 3 and Table 1).

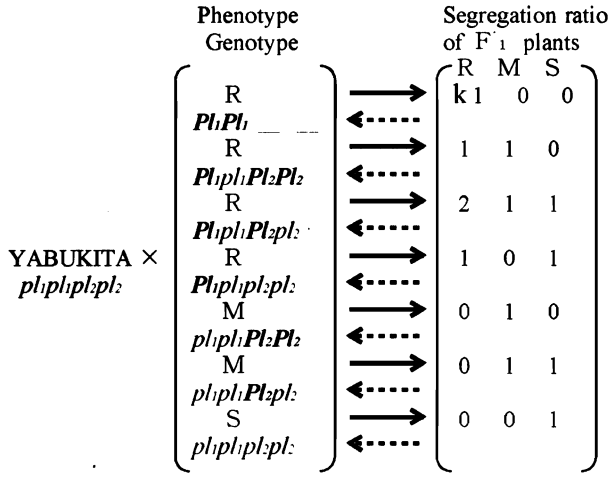

Fig. 3 Genotypes and phenotypes of the tea plants in the resistance to *P. longiseta*

Phenotypes: R (resistance) M (moderately resistance) S (susceptibility)

→ : Theoretical segregation ratio of phenotype in F_1

←--- : Genotypes estimated from the theoretical segregation ratio of phenotypes in F_1

Moreover, the 98 % of the introduced Chinese plants which belong to var. *sinensis* showed the phenotype with a high level of resistance and it was not different from the Assam variety (Fig. 2). However, their genotypes of resistance to tea gray blight were greatly different from the Assam variety, and clear differences were also observed between introduced Chinese plants and Japanese native plants in the Chinese variety. It was observed that the Chinese variety showed wider variations than the Assam variety in genotypes to the disease and it was also observed in the Chinese variety that polymorphism of genotypes in Japanese native plants was wider than that of the introduced Chinese plants. The genotypes of the Assam variety were very

Table 1 Genotype of the tea plants in the resistance to *P. longiseta*

Phenotype / Varieties	Resistance				Moderately resistance		Susceptibility	Total
Genotype	*Pl1Pl1_ _*	*Pl1pl1Pl2Pl2*	*Pl1pl1Pl2pl2*	*Pl1pl1pl2pl2*	*pl1pl1Pl2Pl2*	*pl1pl1Pl2pl2*	*pl1pl1pl2pl2*	
【var.*assamica*】								
Assam plants	96	28	1	4				129
Taiwan wild tea	11							11
【var.*sinensis*】								
Small leaf of Darjeeling	23	6						29
Chinese plants	42	30	2	9	1	5	1	90
Japanese plants	12	29	17	4	8	8		79
Breeding cultivars	3	11	3	9		6	11	43
【Assam Hybrids】	6	18	3	9	4	6	5	51

Genotype of *Pl1Pl1_ _* contains next three genotypes; *Pl1Pl1Pl2Pl2*, *Pl1Pl1Pl2pl2*, *Pl1Pl1pl2pl2*.
Assam plants were collected from India, Sri Lanka, Myanmar, Vietnam and Bangladesh.
Taiwan wild tea means Taiwan mountain wild tea collected from Taiwan.
Small leaf of Dajeeling is a Chinese type plant with small leaves collected from Darjeeling.

simple and 72% of them showed homozygosity for the Pl_1 gene which confers a high level of resistance to tea gray blight. Though the introduced Chinese plants were very similar to the Assam variety in the phenotypes of resistance to the disease, they had wider variations than the Assam variety in the genotype and all seven genotypes were found in them.

The Japanese native plants had widest variation both in genotype and in phenotype among the three groups. In Japanese native plants of the Chinese variety, 83% of the plants showing a high level of resistance had only one Pl_1 gene. The variation of the genotype in the small leaf group of Darjeeling was as narrow as that of Assam plants.

The phenotypes and genotypes of the resistance to tea gray blight of 88 major cultivars in Japan are shown in Table 2. As a high level of resistance gene Pl_1 of the breeding cultivars in Japan derived mainly from introduced foreign tea plants, they contributed to the breeding for resistance to tea gray blight in Japan. The cultivars for 'Sencha', a variety of Japanese green tea, included a high rate of susceptible ones to the disease since the susceptible cultivar, 'Yabukita' was used many times for the breeding materials.

Table 2 Phenotypes and genotypes of 88 cultivars

Phenotype	Genotype	Cultivar
Resistance	$\boldsymbol{Pl_1Pl_1}$ __ __	Benihomare Benihikari Benifuuki Indo Benifuji Inzatsu131 Tadanishiki Fushun Kuritawase Minamisayaka Karabeni Chin-Shin-Oolong San-Cha-Tsi-Lan Chin-Shin-Da-Pan Huang-Gan
	$\boldsymbol{Pl_1pl_1Pl_2Pl_2}$	Benitachiwase Akane Houryoku Satsumabeni Benikaori Yaeho Miyoshi Izumi Takachiho Himemidori Tamamidori Unkai Hoshinomidori Asagiri Yamanami Komakage Z-1 Okuhikari Kanaya-No.15 Makurazaki-No.4 Makurazaki-No.5
	$\boldsymbol{Pl_1pl_1Pl_2pl_2}$	Hatsumomiji Rokuro Koyanishi Shunmei ME-52 Nka-O3 S-6 Makurazaki-No.7 Makurazaki-No.8
	$\boldsymbol{Pl_1pl_1pl_2pl_2}$	Sayamakaori Yamatomidori Surugawase Okumidori Asanoka Minamikaori Satouwase Kanayamidori Ooiwase Shizu-zai-16 Saitama-No.9 Kanaya-No.7 Miyakei-No.2 Makurazaki No.11 Makurazaki-No.13 Makurazaki-No.16 NN-27
Moderately resistance	$pl_1pl_1\boldsymbol{Pl_2Pl_2}$	Makizono-dai-chaju Makurazaki-No.1 Nagasaki-No.2
	$pl_1pl_1\boldsymbol{Pl_2}pl_2$	Yutakamidori Meiryoku Fujimidori Minekaori Kurasawa Yamakai Nka-O-278 Makurazaki-No.18 Makurazaki-No.23 Unryu-cha
Susceptibility	$pl_1pl_1pl_2pl_2$	Yabukita Asatsuyu Saemidori Toyoka Fukumidori Hokumei Sayamamidori Okumusashi Natsumidori Okuyutaka Harumidori Mie-No.260 Kanaya-No.5

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