Breeding of Chestnut Varieties Resistant to Chestnut Gall Wasp, *Dryocosmus kuriphilus* YASUMATSU

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The first breeding program of the chestnut, *Castanea* spp., was started in 1947 at the Horticultural Research Station and planned crossings were completed in 1962. At present, we are conducting breeding works according to the second program.

The aims of the first program were to breed early and mid-season varieties having high productivity, superior quality and good appearance of the nut.

However, in 1941, the chestnut gall wasp, *Dryocosmus kuriphilus* YASUMATSU, was generated in Okayama Prefecture located in the southwestern part of Japan. They spread rapidly at various chestnut growing districts in the 1950's causing great damage to the chestnut trees.

In 1952, a new aim of the development of varieties resistant to the gall wasp was added to the chestnut breeding. Since that time, the character of resistance has been particularly emphasized more and more.

As the result of this program, we commercially introduced four varieties resistant to the gall wasp. Currently, these varieties are the main commercial varieties.

Chestnut gall wasp

In 1941, chestnut trees infested with the gall wasp were found in Okayama Prefecture at first, and the chestnut production was remarkably decreased by the rapid spread of insect in the various growing districts.

Today, this insect is widely distributed all over Japan (Fig. 1), and is also reported to

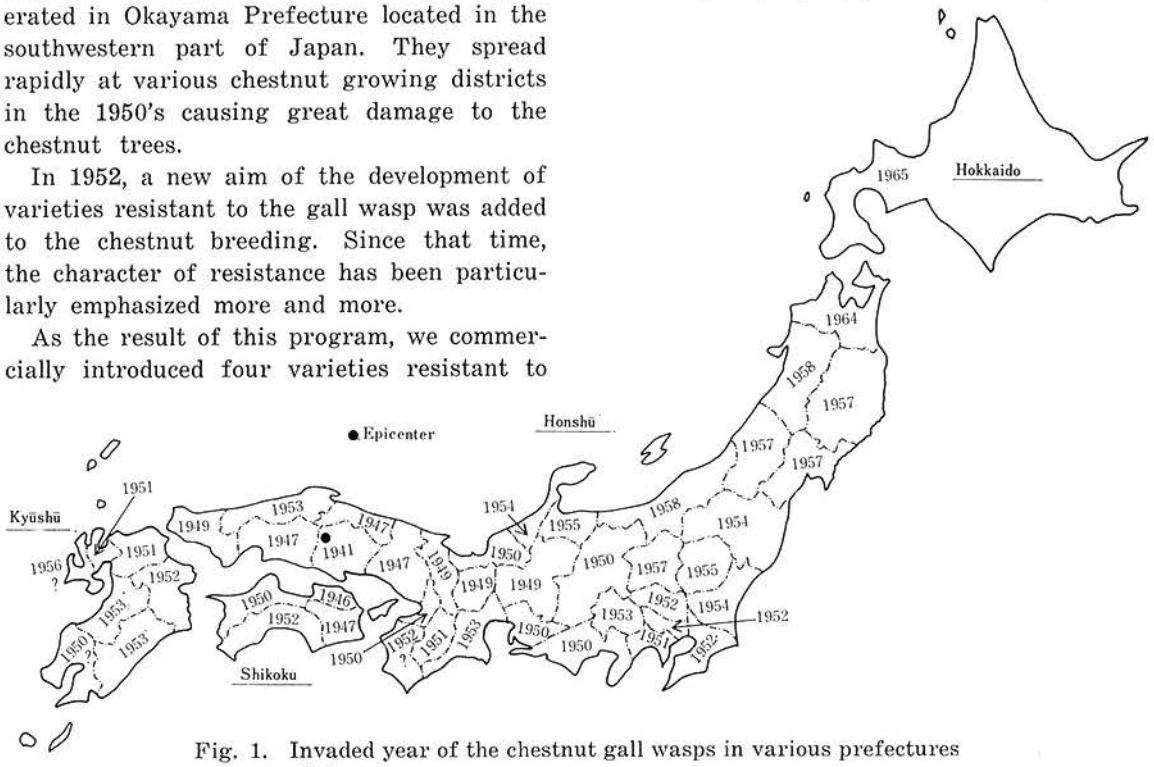

Fig. 1. Invaded year of the chestnut gall wasps in various prefectures of Japan

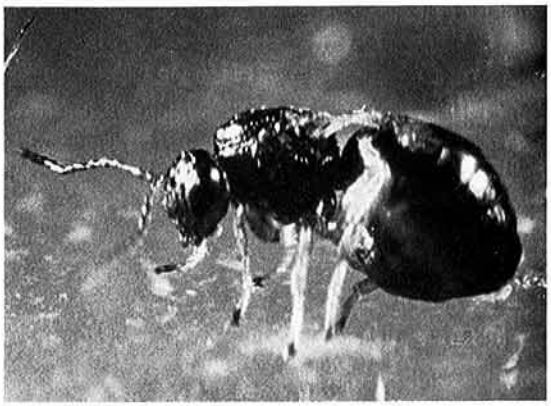

Fig. 2. A female adult of the chestnut gall wasp

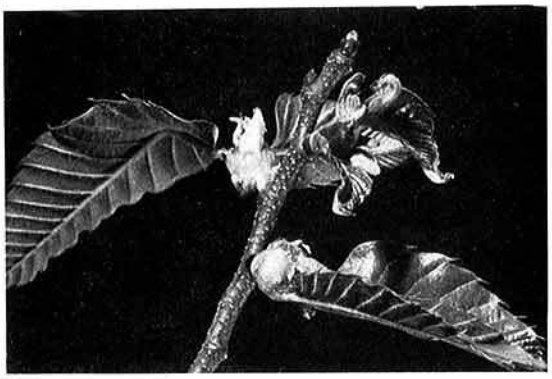

Fig. 3. Galls formed on the branch of two years old chestnut tree

have invaded Korean in 1958.

The adults of these insects are a small gall wasp colored shiny black and their body length is about 3.0 mm (Fig. 2).

They appear from late June to middle July in Hiratsuka, Kanagawa Prefecture, and lay eggs in the dormant buds formed on the current shoots. The eggs hatch in August and the larvae invade the bud tissue immediately, passing the winter in the tissue with larva form.

In spring, they grow in the tissue and make a gall on the parasitic bud in April (Fig. 3). Consequently, the nut production is remarkably decreased by the check of current shoot growth.

Selection and breeding of chestnut resistant to the gall wasp

Extensive studies have been conducted by a number of entomologists to control the wasp through various ways including application of chemicals and natural enemies, but all their efforts were in vain.

On the other hand, some varieties among the chestnut cultivars were found to be apparently resistant to the wasp in the field observations. The cultivars could be classified

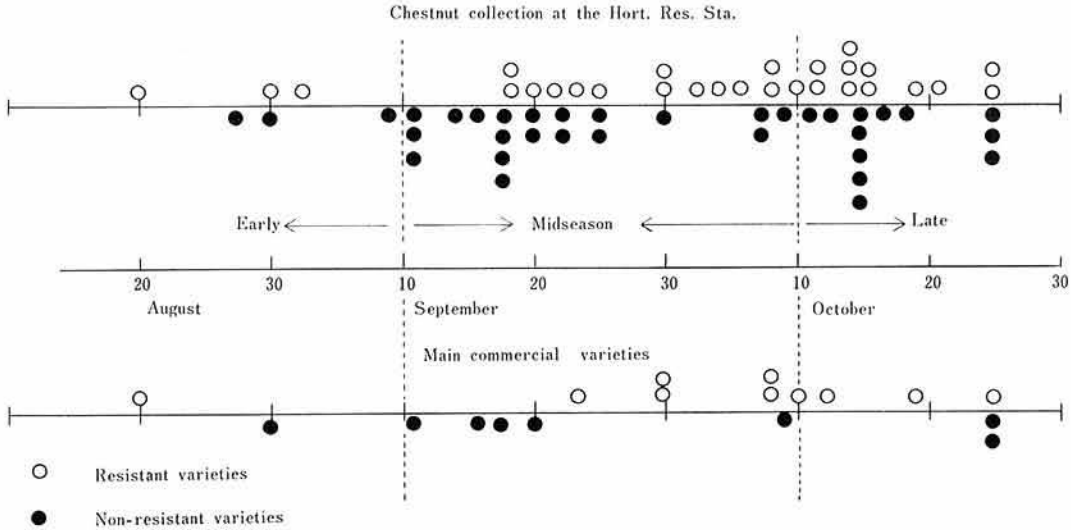

Fig. 4. Ripening periods of the resistant and non-resistant varieties to chestnut gall wasp (KAJIURA, 1955)

into two groups, the resistant and the non-resistant. Non-resistant varieties disappeared from commercial growing because of their severe infestation.

The culturists emphasized the need of breeding of the resistant varieties that ripen in September since many of the varieties ripening during that time had been severely damaged by the insect. This was a serious problem because the nuts are in the greatest demand in September (Fig. 4).

The first program of chestnut breeding was conducted from 1947 to 1962 at this station. The crossings of 139 combinations were done between 18 varieties as female parent and 28 varieties as pollen parent.

A total of 3,597 hybrids from 3,946 obtained seedlings were grown in the experimental fields and subjected to tests. Thirteen individuals resistant to the wasp were selected and released in 1956 in order to study the general characters of the tree, productivity and resistance to the gall wasp and to blight fungus at 40 prefectural experiment stations. Four individuals were selected from this cooperative tests continued until 1967.

These individuals were named 'Tanzawa' (chestnut Nōrin No. 1), 'Ibuki' (chestnut Nōrin No. 2) and 'Tsukuba' (chestnut Nōrin No. 3) in 1959, and 'Ishizuchi' (chestnut

Table 1. Development of varieties bred in the Horticultural Research Station

Registered No.[a]	Variety	Parentage	Crossed year	Year of the first stage selection at the Hort. Res. Sta.	Test year[b] for general characteristics	Named and released year
Chestnut Nōrin No. 1	Tanzawa	Otomune × Taishōwase	1949	1955	1956～1958	1959
Chestnut Nōrin No. 2	Ibuki	Ginyose × Toyotamawase	1947	1955	1956～1958	1959
Chestnut Nōrin No. 3	Tsukuba	Ganne × Hayadama	1949	1955	1956～1958	1959
Chestnut Nōrin No. 4	Ishizuchi	Ganne × Kasaharawase	1948	1955	1956～1967	1968

[a] Registered No. at the Ministry of Agriculture and Forestry, Japan
[b] Cooperative tests were made at 40 places of various prefectural experiment stations

Table 2. Characteritics of Tanzawa, Ibuki, Tsukuba and Ishizuchi bred in the Hort. Res. Sta.

Variety	Tree	Nut
Tanzawa	Vigor medium, not timber-type, blooms from early to middle June	Large, average weight 20 g, shell light brown, pellicle not easily separable, quality moderate, ripense from early September
Ibuki	Vigor medium, not timber-type, blooms early to late June, bears heavily	Large, average weight 20 g, shell dark brown, pellicle not easily separable, quality good, ripens from early to middle September
Tsukuba	Upright, vigorous, not timber-type, blooms from early to late June, bears heavily	Large, average weight 20 g, shell reddish brown, pellicle not easily separable, quality good, keeping quality superior than Tanzawa and Ibuki varieties
Ishizuchi	Vigor medium, not timber-type, blooms from middle June to early July, bear heavily	Very large, average weight 23 g, shell shiny brown, quality good, keeping quality good, ripens from early to middle October

Nōrin No. 4) in 1968, all being introduced commercially (Tables 1 and 2). No sooner had they been commercially introduced than they were planted in commercial orchards. Consequently, the acreage of chestnut as orchard trees has been remarkably increased (Fig. 5).

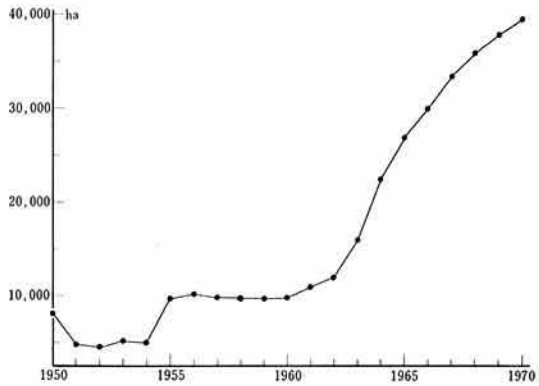

Fig. 5. Fluctuations of the chestnut growing acreage in Japan

Resistant mechanism and heredity

Fukuda and Okudai (1951) observed the egg-laying preference of the wasp on the resistant and non-resistant varieties. From this observation, they concluded that the gall wasp could not have a non-preference of egg-laying between the resistant and non-resistant varieties.

Thereafter, Okudai (1956) made a microscopic observation on the parasitic buds collected seasonally from both the resistant variety, 'Ginyose', and the non-resistant variety, 'Taishōwase'. From the result, he found that the hatched larva invading the bud of the non-resistant variety, 'Taishōwase', grew up to an adult normally in it. But, with the resistant variety, 'Ginyose', the tissue surrounding the living chamber of the larva induces the necrosis.

The larva, consequently, died from juvenile stage to pupa stage in the parasitic bud. From these results, he concluded that the resistance to the wasp of the resistant variety is of a real resistant nature.

Recently, Torikata and Matsui (1965), and Matsui and Torikata (1968) conducted a chemical analysis to clarify the biochemical differences between the resistant and non-resistant varieties.

They pointed out that the contents of catechol tannin and leucoanthocyanidin in the bark of resistant varieties were higher than those of the non-resistant varieties and that the pyrogallol tannin content in the resistant varieties was lower than that of the non-resistant varieties.

Furthermore, they obtained results that the correlation coefficients between the resistance grade of the varieties to the gall wasp and catechol tannin content, and pyrogallol tannin content were $+0.782^{**}$ and -0.476^{*}, respectively.

Consequently, they guessed that the catechol tannin may be one of the factors affecting the resistance to the gall wasp. However, the mechanism of resistance is not yet made clear in every respect.

Regarding the inheritance of the resistant character, Kajiura (1955) reported the rate of resistant seedlings in the hybrids obtanied from various crosses. He showed that the high percentage of resistant seedlings appeared when the resistant variety was used as a female or male parent for the cross (Table 3).

Especially, almost all of the F_1 hybrids obtained from the cross of 'Ginyose' used as a female or male parent were resistant seedlings. The performance of the cross A in Table 3, suggested that the 'Ginyose' variety has a homozygous dominant factor to the resistance, but that 22.4 per cent of the resistant seedlings obtained from the cross E showed the existence of two or more factors concerned with the resistance.

Though these results could not be analyzed sufficiently yet, the data proved to be a matter of great importance for our breeding works. They showed that resistant varieties could be obtained without much difficulty since the horticultural varieties are propagated by

Table 3. Segregation of resistant seedlings to the chestnut gall wasp in the hybrid seedlings derived from various crosses (KAJIURA, 1955)

Combination*	Hybrid seedlings	Resistant seedlings	Non-resistant seedlings	Percentage of non-resistant seedlings
A	86	86	0	0
B	258	257	1	0.4
C	170	148	22	12.9
D	302	231	71	23.5
E	170	38	132	77.6

*A : Ginyose × Resistant varieties except Ginyose
B : Ginyose × Non-resistant varieties
C : Resistant varieties except Ginyose × Resistant varieties except Ginyose
D : Resistant varieties except Ginyose × Non-resistant varieties
E : Non-resistant varieties × Non-resistant varieties

grafting.

Thanks to these studies, the injury of chestnut trees by the gall wasp has been almost completely controlled by growing resistant varieties.

Parasitic variation of the chestnut gall wasp

Recently, however, the gall wasp is recognized to infest the resistant varieties in the commercial fields in several chestnut growing districts. Besides, the population density of the wasp as well as the infested area has a trend to increase.

In order to clarify this phenomenon, the author has conducted experiments from the viewpoints of the parasitic variation of the gall wasp.

The experiments were carried out: (a) to make a gall by the inoculation of the wasp collected from different hosts of various cultivated varieties in the field at the Horticultural Research Station, and (b) to study the variation of polyphenol oxidase and peroxidase isozymes in the wasps collected from various hosts.

The inoculation of the wasp collected from the resistant host, 'Tanzawa', caused gall formation on both the resistant and non-resistant varieties tested.

However, the gall wasp from the non-resistant hosts, 'Miyagawa-85' and 'Shibaguri', induced gall formation only on the non-resistant varieties and not on the resistant varieties (Table 4).

Table 4. Rate of gall formation by the inoculation of chestnut gall wasps collected from various hosts

Host variety of the chestnut gall wasp	Resistant variety			Non-resitant variety		
	Tanzawa	Otomune	Tsukuba	Hayadama	Yamewae	Nakatetanba
Tanzawa*	41.2%	76.9%	41.7%	95.5%	97.2%	89.5%
	7/17***	10/13	5/12	21/22	35/36	17/19
Miyagawa-85**	0	0	0	50.0	100	100
	0/5	0/19	0/5	2/4	27/27	10/10
Shibaguri**	0	0	0	50.0	100	27.8
	0/14	0/15	0/4	5/10	15/15	5/18

* TANZAWA is the resistant variety to the chestnut gall wasp
** Miyagawa-85 and Shibaguri are the non-resistant varieties to the chestnut gall wasp
*** Galls/Buds of inoculation

No significant difference of polyphenol oxidase isozyme was observed between the insects obtained from the resistant host, 'Ibuki', and those from the non-resistant host, 'Miyagawa-85'.

But there was a remarkable difference of peroxidase isozymes among the insects collected from various hosts. The insects obtained from the resistant hosts, 'Ibuki' and 'Tsukuba', had a kind of peroxidase isozyme moving toward cathode and two kinds of the isozymes moving toward anode.

On the other hand, the insects from the non-resistant host, 'Miyagawa-85', had two kinds of isozymes moving only toward anode (Fig. 6).

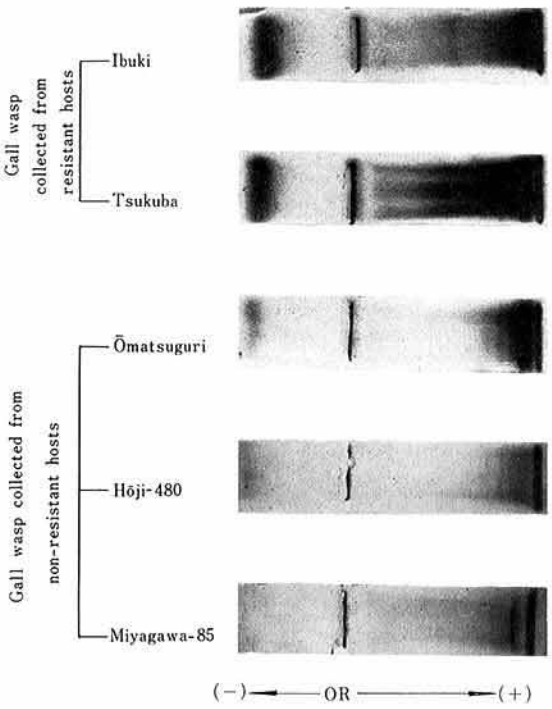

Fig. 6. Peroxidase zymograms of the chestnut gall wasp collected from various hosts

The wasp collected from galls formed on 'Mayagawa-85' by means of the inoculation of the wasp obtained from 'Ibuki' showed the same zymograms as the insects collected directly from 'Ibuki'.

Therefore, it is obvious that the differences of the peroxidase isozymes between the insects from the resistant and non-resistant hosts are induced genetically.

The wasp obtained from 'Omatsuguri' and 'Hōji-480', both being non-resistant hosts, also had the isozyme moving toward cathode, but the activity of this isozyme was inferior to that of the insects from resistant hosts. Therefore, it seems that some wasps having isozyme moving toward cathode may exist in the gall of these non-resistant hosts.

From the results obtained above, it is concluded that the injury on the resistant varieties has been caused by the infestation of the gall wasp having a new parasitic ability, and that the gall wasp has been differentiated into at least two groups in the population.

With the development of such a parasitic ability of this insect, the breeding of chestnut varieties resistant to the gall wasp has now encountered new problems.

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