# Development of a Novel Formulation: Pluto<sup>®</sup> MC to Control Mulberry Scale on Tea

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Pluto<sup>®</sup> MC is a new concept insecticide which was developed by Sumitomo Chemical Co., Ltd. to control mulberry scale on tea during winter dormancy (from January to March). This formulation is a self-burst type of micro capsule formulation containing 9% pyriproxyfen to reduce the risk of drift when it is sprayed in the tea field.

Dormant spray of this formulation can show good control efficacy against the mulberry scale for a longterm period and can greatly reduce the risk of toxicity to silkworms at the same time. Also, it is promising for farmers as a new labor-saving technique. Moreover, this insecticide is suitable for integrated pest management (IPM) programs.

In this report, the development of Pluto<sup>®</sup> MC is described focusing on basic performance against the mulberry scale, formulation design, optimum application timing and efficacies.

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### Introduction

Pluto<sup>®</sup> MC is a new insecticide formulation developed by Sumitomo Chemical with the insect growth regulator (IGR) pyriproxyfen as the active ingredient. Pyriproxyfen is a compound with a long track record that has been registered and sold in various countries around the world starting in the Middle East in 1988 under trademarks such as Knack<sup>®</sup>, Admiral<sup>®</sup> and Esteem<sup>®</sup> as a pest control for whiteflies, scale insects and thrips.<sup>1)</sup> In particular, since it has an extremely high level of activity and exhibits a superior control efficacy for whiteflies and scale insects, it has been given high marks at agricultural sites in various countries.

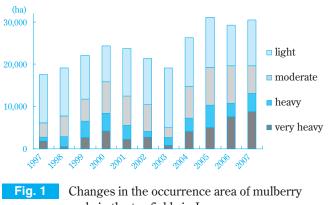
Since pyriproxyfen strongly affects silkworms, domestic development has been delayed because of the necessity for taking the sericultural industry into consideration, but in 1997 the Lano<sup>®</sup> emulsifiable concentrate for thrips and whiteflies and the Lano<sup>®</sup> tape for whiteflies on vegetables in greenhouses were developed and marketed.<sup>2)</sup> In particular, in addition to getting rid of concerns about the risk of toxicity to silkworms by establishing a completely new landmark application technology that reduces the work in application with a non-spreading application, Lano<sup>®</sup> tape has had a timely coincidence with the demand for labor saving pesticide technology that has been increasing at agricultural sites in recent years. As a result, Lano<sup>®</sup> tape has penetrated widely into agricultural sites where there is institutional cultivation as a control agent for whiteflies, and it plays a role as a key control resource in control technologies for IPM.

We have now developed the new formulation Pluto<sup>®</sup> MC for mulberry scale (*Pseudaulacapsis pentagona*, also known as white peach scale), which is an insect harmful to tea and which is difficult to control, as a second shot at a labor-saving technique using pyriproxyfen in Japan. In this paper, we will give an introduction to features such as the background and history of development, mechanism of action, design of formulation, practical control efficacy, and the effects on natural predators and useful insects.

#### **Background to Development**

In recent years, there has been a trend towards increasing occurrence of and damage by scale insects in Japan, and in particular, there has been an escalation in the problem of mulberry scale in tea (Fig. 1).<sup>3)</sup> This harmful insect produces three to four generations per year, and parasitizes and sucks on the branches of tea plants. Repeated occurrences not only make the

growth of tea shoots poor, but also cause severe damage where the tea plants wither in the worst cases. This harmful insect is one that is very difficult to prevent among tea pests, and the following may be cited as causes for this. (1) Since the eggs and adults are covered with a scaly substance, they are shielded by the scaly substance even if insecticides are applied, and it is difficult to apply liquid insecticides to the bodies of the insects. (2) The suitable period for control for existing insecticides is limited to several days after the larvae hatch and before the formation of the scale, but various conditions even within the same tea field are not uniform when the larvae hatch, so it is difficult for it always to be the optimal time for control.



scale in the tea fields in Japan. [Data from reference 3)]

(3) Since the parasitism is in the branches in the crown of the tea plants, it is difficult for the insecticide to sufficiently reach the parasitism from the thick layer of leaves during the suitable period for control.
(4) Among the limited control agents, there have been reports in recent years of reduced sensitivity to existing insecticides in some regions.<sup>4)</sup>

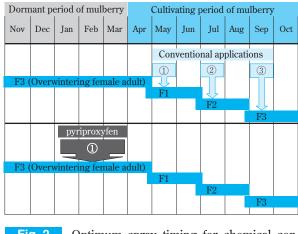
Furthermore, a large volume of water for application of 1000 L/10a is required to obtain a sufficient control efficacy in dealing with the situation described in (3) above, and this treatment requires a large amount of time and effort. In addition, the time suitable for preventing the first generation is the busiest period for first tea-picking, and the second generation coincides with the extreme heat of midsummer in July. This places a great burden on tea farmers.

Therefore, we carried out initial investigations into whether pyriproxyfen, which has a good record overseas as a insecticide for scale insects, is sufficiently active on mulberry scale, and from those results decided to investigate the possibilities of developing a laborsaving control technique. Furthermore, because of the strong fear of this insecticide having adverse effects on the silkworm industry, we thought it was necessary to construct a new control technique to avoid unforeseen accidents involving silkworms.

# Possibilities for Control of Mulberry Scale through Dormant Spray Application

The best measure for preventing silkworm accidents with pyriproxyfen is not allowing the insecticide to scatter to the mulberry leaves.

The mulberry, which is the food for silkworms, is a deciduous shrub, so it is in a dormant state during the winter without any open leaves. In other words, even if there are incidents where the insecticide hits mulberry trees because of unforeseen drift, there can be a large reduction in the possibilities of incidents of silkworm toxicity occurring if it is a dormant spray application. Furthermore, if it is winter, which is the off season for farmers who cultivate tea, we thought that there could also be a great reduction in the burden on the tea farmers (**Fig. 2**). However, in such a case, the most important question to arise was whether or not pyriproxyfen would have the performance to give sufficient control for the target insects over the summer season after a winter application.





Optimum spray timing for chemical control of mulberry scale

# 1. Activity on mulberry scale during different growth stages

First of all, we evaluated the activity of pyriproxyfen on mulberry scale by growth stage. Our results showed that it exhibited a high level of ovicidal activity with immersion treatment of the eggs (**Table 1**). In addition, in egg inoculation tests on treated plants during the period of molt from the first instar to the second instar, it was clear that there were primarily mortality (**Table 2**). Most of the dead insects had their growth

Table 1	Ovicidal	activity	of	pyriproxyfen	against
	mulberry	v scale			

	Conc.	9DAT*		
Chemicals		# of settled Control		
	(ppm)	1 <sup>st</sup> nymphae	efficacy	
	90	3.7	99%	
pyriproxyfen	9	32.3	91%	
untreated	_	372.5	0%	

Treatment methods : Egg immersion (5 sec.)

\* : Days After Treatment

Table 2	Molt inhibiting activity of pyriproxyfen
	against 1st nymphae of mulberry scale

Chemicals	Conc.	Molt inhibiting	Control
enemicais	(ppm)	rate	efficacy
pyriproxyfen	90	99%	98%
	9	84%	82%
untreated	_	6%	_

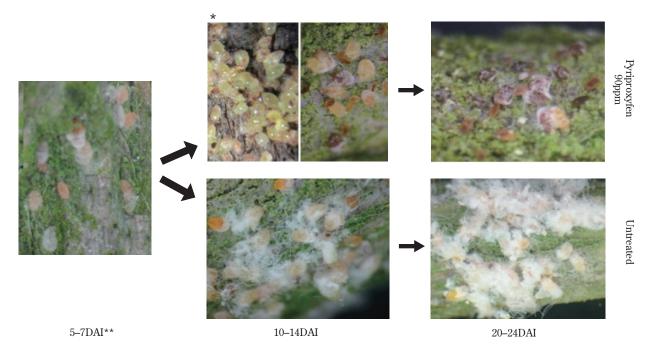
Treatment method : Foliar spray. Eggs wrapped with KIMWIPES were hanged on the treated branches to allow newly hatched nymphs to move to the branches.

stopped in a state where the scaly substance had clearly been released from the back and they had turned brown and died afterwards. In addition, we also found cases where the bodies of the insects exhibited swelling during the period where the growth stopped (Fig. 3). However, when we applied pyriproxyfen starting in the second instar, we did not find inhibitory activity for the growth to adult insects in females (Table 3). Furthermore, when the insecticide was applied to overwintering female adult insects parasitizing tea plants in winter (March), the number of incubated eggs was somewhat lower than for untreated insects, but the hatching rates for the treated group and untreated group were almost the same (Table 4). We presume the reason that the hatching rate was no lower for the treated group was that the eggs were protected by the scale of the females so that the active ingredient did not reach them.

# Table 3Activity of pyriproxyfen against 2nd instar<br/>female nymphae of mulberry scale

Chemicals	Conc.	Growth inhibition rate to adult
pyriproxyfen	(ppm) 90	0%
untreated	00	10%

Treatment method : Pumpkin fruit immersion (5 sec.)



\* : Mie Prefecture Agricultural Research Institute

\*\* : Days After egg Inoculation

Fig. 3 Symptoms of pyriproxyfen on nymphae of mulberry scale

	Conc.	Rate of	Mean # of eggs	Growth rate from egg
Chemicals		oviposition	per female adult ±SE	to 2 <sup>nd</sup> instar larva
	(ppm)	female adult		±SE
pyriproxyfen	90	90%	$49.3\pm10.0$	$28.4\%\pm7.05$
untreated		100%	$61.2 \pm 10.0$	$24.5\% \pm 6.33$

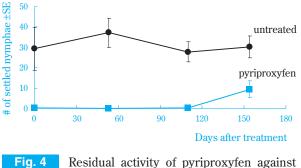
Table 4	Activity of pyriproxyfen against young female
	adult of mulberry scale

Treatment methods : Mulberry scale infested fruit immersion (5 sec.)

From the results above, we assume that there is no effect on overwintering female insects when winter applications of pyriproxyfen are made, and the control efficacy is first expressed when the larvae that are hatched subsequently come into contact with the pyriproxyfen.

#### 2. Residual effect on mulberry scale

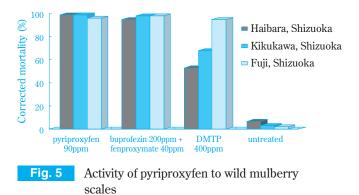
From the results above, it became necessary to confirm what the length of the period of residual effectiveness of the winter application of pyriproxyfen on mulberry scale would be for showing a control efficacy. To achieve this, pyriproxyfen was applied to potted tea plants. Eggs were periodically inoculated while being kept in an open field, and evaluations were carried out. As a result, it was confirmed that the effect of controlling high densities continued for a long period of time of from 100 to 150 days after treatment (**Fig. 4**).



. 4 Residual activity of pyriproxyfen against mulberry scale

# 3. Activity on lines of mulberry scale with reduced sensitivity to existing insecticides

In recent years, some regions have reported a reduction in the sensitivity of mulberry scale to existing insecticides.<sup>4)</sup> Therefore, we conducted susceptibility monitoring on pyriproxyfen using a group of individuals collected from tea fields in Shizuoka Prefecture. Specifically, this was conducted using a test method where cut branches of tea were treated with an application of a prescribed concentration of this agent, and after air drying, eggs were inoculated. Larval parasitism was checked 14 days after inoculation. As a result, pyriproxyfen exhibited a level of insecticidal activity on the scales collected from Haibara and Kikukawa, where there was reduced sensitivity to DMTP, as high as an insecticide susceptible strain, and the cross resistance with existing insecticides was determined to be extremely low (**Fig. 5**).



From the results above, it was determined that practical control efficacy could be expected for mulberry scale prevention using one winter treatment with pyriproxyfen.

#### **Fundamental study on MC Formulation**

#### 1. Less drift formulation

Typically, a preparation is diluted with water for liquid application, and applied in the form of a spray using equipment such as a sprayer, so it is easy for drift to occur. Not only does drift cause losses in the liquid insecticide being applied, but also, when the agricultural chemical reaches locations other than the agricultural fields targeted for prevention, there is a danger of undesirable residues accumulating and chemical damage to crops on which it is not targeted.<sup>5)</sup>

While winter application was considered as a countermeasure for preventing drift of this insecticide as described in the previous section, we thought in terms of implementing further drift countermeasures in a functional formulation, and set a formulation with low drift as our first development goal.

Typically, it has been said that the spray solution may be made so that it will not drift easily by making large droplets in the spray solution, which increases the sedimentation properties in the air. Therefore, we first considered the influence of spray drop size as a main factor (**Fig. 6**).<sup>6)</sup>

$$D = 47 \ \frac{D_N}{V} \ (\frac{\sigma}{\gamma})^{0.25} \ \frac{\gamma}{\gamma_a}^{0.25} \ \sqrt{g} \ (1 + 3.31 \ \frac{\mu\sqrt{g}}{\sqrt{\sigma} \cdot \gamma \cdot D_N} \ )$$

DN; Diameter of the nozzle,  $\sigma$ ; Surface tension,  $\mu$ ; Viscosity g; Gravity acceleration, V; Spray speed,  $\gamma_a$ ; Specific gravity of the air,  $\gamma$ ; Specific gravity of the liquid

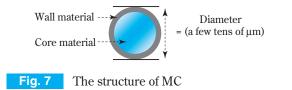
Fig. 6 Diameter (D) of the droplet after spraying

In the equation in Fig. 6, factors that can increase D are surface tension and viscosity. Since this insecticide is diluted with a large amount of water when used, we assumed that there would be very little difference in viscosity between formulations before and after the dilution, so we focused on surface tension.

Here we compared the physical properties of diluted solutions of an emulsifiable concentrate (EC) and a microcapsule (MC), which are representative of the products sold by Sumitomo Chemical (**Table 5**). A diluted MC solution generally has greater surface tension than a diluted EC solution, so from the equation in Fig. 6, the droplet diameter after spraying is larger, suggesting that it is a formulation that makes drift more difficult (**Fig. 7**).

Table 5	Physicochemical properties of emulsifiable
	concentrate (EC) and microcapsule (MC)

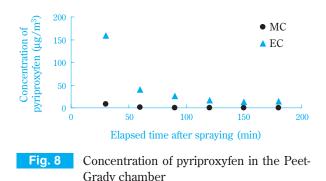
Formulations	EC	МС	
Surface tension of the	Low	High	
diluted formulation	Low		
Diameter of the droplet	Small	Big	
after spraying	Sillan		



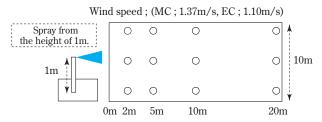
To verify the ideas above, we carried out the following tests for residual pyriproxyfen concentration in the air after we sprayed the diluted formulations.

We sprayed a diluted MC solution and a diluted EC solution containing pyriproxyfen in a Peet-Grady Chamber  $(1.8m \times 1.8m \times 1.8m = 5.8m^3)$ , and after the prescribed period of time, we compared the pyriprox-

yfen concentrations in the air (**Fig. 8**). The concentration of the sprayed MC in the air decreased more rapidly than the sprayed EC. This proved our hypothesis.



Next, we sprayed the diluted MC solution and the diluted EC solution containing pyriproxyfen outdoors (at the Sumitomo Chemical, Kasai Experimental Farm) and measured the pyriproxyfen concentrations at various sites (at the circles in the diagram) two hours after spraying (**Fig. 9**).



Nozzle type : Compact nozzle 2 heads Pressure : 15kg/cm<sup>2</sup> Dilution ratio : 1000times Spray volume : 20L

Distance from the spray place (m)	2	5	10	20
MC (ppm)	56.6	2.5	0.7	No detection
EC (ppm)	69.4	12.2	1.9	No detection

Fig. 9 Test method and the result of the drift study

These results suggested that even in outdoor tests, the sedimentation properties in the air were superior for the MC solution.

We selected MC as the lower drift formulation from these verification tests.

#### 2. Self-burst MC development

As was discussed earlier, there was a suggestion of the possibilities for pyriproxyfen having a distinct efficacy for mulberry scale with winter spraying (Table 1-4, Fig. 3). In other words, if the pyriproxyfen adheres to the tea plants after the formulation is sprayed, we can assume that the effect will continue for four to five months or more. However, the MC formulation is covered with a wall material, so the pyriproxyfen itself does not contact the tea plant directly. Therefore, the second developmental goal for this functional formulation was to give it the new function of securely bringing pyriproxyfen into contact with the tea plants after the MC wall material bursts.

From the two development goals mentioned previously, we considered that it was necessary to develop an MC formulation with a new "self-burst" function that would maintain the MC configuration in the spray solution, make the wall material rapidly burst after the MCs adhere to the surface of the tea branches and make the core material contact the tea plants (**Fig. 10**).

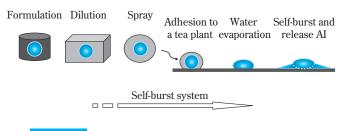
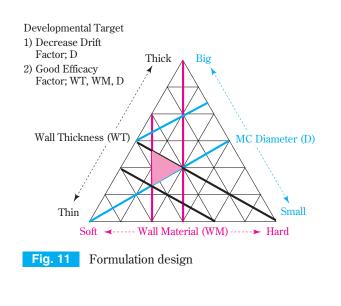


Fig. 10 Concept of the Self-burst MC

Since we have researched, developed and sold many MC formulations up to now at Sumitomo Chemical<sup>7</sup>, we made use of our prior knowledge in this project in order to further reduce the drift in the MC formulations and to bring out the efficacy to the maximum extent, and we made careful selection of the MC diameter, wall thickness and wall material as is shown in **Fig. 11**. The parameters showing the hardness and elasticity also had to be optimized. When the diluting water evaporated on the plants after the Pluto<sup>®</sup> MC which was optimized based on these ideas was diluted with water and sprayed, the wall material burst and the active ingredient was released. A microscopic photograph of this is shown in **Fig. 12**.

On the other hand, the fact that the active ingredient is not released in the diluted solution and that it is present as MCs in a stable form can be confirmed by the fact that the amount of the active ingredient outside of the wall material before and after dilution does not change (**Table 6**).



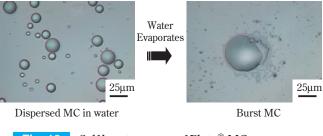


Fig. 12 Self-burst process of Pluto<sup>®</sup> MC

Table 6

Pyriproxyfen content outside of MC in a dilute solution\* (w/w%)

Conditions	Content (w/w%)
Initial	0.0002
After 24h	0.0002

\* Dilute 1 g of Pluto® MC with 999 mL of deionized water.

In addition, as this formulation was designed to have a low viscosity and a small amount of foaming, it makes for a formulation design with superior handling qualities, and the storage stability is also very good.

From the results of these investigations, we were able to bring to fruition the new self-burst MC formulation of Pluto<sup>®</sup> MC which has (1) less drift, (2) quick release properties and (3) superior physical stability.

# **Evaluation of Practical Control Efficacy of MC** Formulation

The overwintering dormant period where the leaves have fallen from mulberry trees is the period from November to the following March. As a result of evaluating residual properties using a pot test for control of occurrences up through the second generation in July with treatment during this period, it was estimated that treatment is necessary in February to March, which is approximately 100 to 150 days prior to the second generation (Fig. 4).

However, we conducted field evaluations of the residual activity at various treatment times including investigations into the possibility of expanding the period of use to spraying in January, with the goal of improving usability.

As a result, for all of the treatment periods in January, February and March, this formulation did not only control the density of male cocoons of the second generation in the field but also showed excellent long lasting effects down to the third generation (**Table 7**). It was clear that the density control effect covered a

longer period than the results of the residual property tests in potted tea plants. In the evaluation of the control efficacy through the third generation, it is easy for the estimated drop in density to come from natural enemies, and there are many cases of tests where it is difficult to make evaluations accurately. Therefore, we monitored the occurrence of parasitic wasps, which are the main natural enemy of mulberry scale using yellow sticky traps, and then carried out evaluations under control conditions where pyrethroids were sprayed at the appropriate time to eradicate these natural enemies. Even in this case, this formulation exhibited a sufficient control efficacy for the third generation (Table 8). In addition, even under conditions where occurrences of the scale increased toward autumn, we obtained cases where a high level of control efficacy was exhibited through the third generation (Table 9).

 Table 7
 Control efficacy of pyriproxyfen (Pluto<sup>®</sup> MC) to mulberry scale by dormant spray

1 Fukuoka Agricultural Research Center (2005, Yame, Fukuoka)

Samples	Conc.	Spray	Appl.		Infestation index $^{j}$	
Samples	(ppm)	Volume	Date	$1^{st} G^{\#}(6/15)^{\dagger}$	2nd G(8/11)	3 <sup>rd</sup> G(10/6)
pyriproxyfen MC	90	1000L/10a	1/28	0.49	0.02	0.00
pyriproxyfen MC	90	1000L/10a	3/14	0.48	0.02	0.00
buprofezin + fenpyroximate FL	200 + 40	1000L/10a	5/20	0.29	0.38	1.17
untreated				1.18	0.62	1.30

2 Nagasaki Agriculture and Forestry Experiment Station (2005, Higashisonogi, Nagasaki)

Samples	Conc.	Spray	Appl.		Infestation index	
	(ppm)	Volume	Date	1 <sup>st</sup> G(6/17)	2nd G(8/19)	3 <sup>rd</sup> G(10/13)
pyriproxyfen MC	90	1000L/10a	2/6	0.97	0.10	0.13
pyriproxyfen MC	90	1000L/10a	3/14	0.11	0.27	0.30
DMTP EC	400	1000L/10a	5/27	0.43	0.03	0.23
untreated				2.10	1.57	1.43

J: The infestation index was calculated as an average of male cocoons infestations scores for 10 places per plot.

No infestation was scored as 0, slightly as 1, light as 2, moderate as 3, heavy as 4. (3 reps)

#: G; generation in a year. †: Observation date.

#### Table 8 Control efficacy of pyriproxyfen (Pluto<sup>®</sup> MC) to mulberry scale under natural enemies elimination\*

Mie Prefecture Agricultural Research Institute (2006, Kameyama, Mie)

				8		
Samples	Conc.	Spray	Appl.		Infestation index $^{\int}$	
	(ppm)	Volume	Date	1 <sup>st</sup> G <sup>#</sup> (6/15) <sup>†</sup>	2nd G(8/11)	3 <sup>rd</sup> G(10/6)
pyriproxyfen MC	90	1000L/10a	3/9	0.33	0.37	0.47
buprofezin + fenpyroximate FL	200 + 40	1000L/10a	5/31, 7/31	0.27	0.17	0.53
untreated				1.67	1.10	1.53

\*: Permethrin EC (5/24, 7/26, 9/20) and cypermethrin WP (7/6) were applied to eliminate parasitic wasps in the test field.

J: The infestation index was calculated as an average of male cocoons infestations scores for 10 places per plot.

No infestation was scored as 0, light as 1, moderate as 2, heavy as 3. (3 reps)

#: G; generation in a year. †: Observation date.

		0	·	2	· · · ·	
Samples	Conc.	Spray	Appl.		Infestation index $^{\int}$	
Sampies	(ppm)	Volume	Date	$1^{st} G^{\#}(6/15)^{\dagger}$	2nd G(8/11)	3 <sup>rd</sup> G(10/6)
pyriproxyfen MC	90	1000L/10a	3/24	0.45	0.28	0.18
DMTP EC	400	1000L/10a	5/29	0.40	2.45	2.30
untreated				1.25	2.57	2.45

 Table 9
 Control efficacy of pyriproxyfen (Pluto<sup>®</sup> MC) to mulberry scale under heavy infestation

National Institute of Vegetable and Tea Science Kanaya Tea Research Station (2006, Shimada, Shizuoka)

J: The infestation index was calculated as an average of male cocoons infestations scores for 10 places per plot.

No infestation was scored as 0, light as 1, moderate as 2, heavy as 3. (3 reps)

#: G; generation in a year. †: Observation date.

However, when the results on the tests were analyzed in detail, we found that there was not complete control of the scale over the long term from the effects of this preparation alone. Even though there was control to a density lower than areas without treatment to the third generation in areas treated with this formulation in field test cases where occurrences of parasitic wasps were controlled, we found there was a trend toward density recovery starting in the second generation (Table 8). The causes of this are thought to be that (1) a certain amount of unevenness of spray cannot be avoided even if the spray treatment is done carefully; (2) the growth of tea plants increases remarkably starting in April, and there is a gradual increase in branches that have none of the formulation adhering to them; and (3) from the results of tests of residual properties, the control efficacy for scale was observed to diminish starting 150 days after treatment (Fig. 4). The control efficacy on the third generation is presumed to be the results of the effect of density control up to the second generation with the addition of the effect of density control by natural enemies.

In addition, from the results of indoor insecticidal activity tests, it can be assumed that there is a possibility of no reduction in first generation larvae that are incubated in May, and sucking damage occurs to the first tea crop when the parasitic density of these insects is high in the autumn, even with winter treatment with this formulation. Therefore, it can be assumed that there is a need for implementing supplemental control for the third generation with another insecticide according to the state of occurrence in autumn.

From the results above, we determined that longlasting control effects on mulberry scale are surely possible through the second generation in July from one treatment with this formulation in winter (during January through March), and further on the third generation in September, depending on the field conditions. Moreover, since this formulation does not have a systemic action, the main point for obtaining stable control efficacy is even spraying. Therefore, it is vital that the sprayer have a special nozzle which is suitable for mulberry scale, and that spray treatment be carried out with a suitable amount of water.<sup>8)</sup> 1000 L/10a is the optimal content of registration for the amount of water in the spray, and it has been reported that if the water content is too much lower than this, uneven spray application arises easily, and if it is too much more than this, the operation is a waste.<sup>9)</sup>

In addition, there is a need to avoid mixed use of other formulations and functional spreading agents that can affect the film structure after use of the Pluto<sup>®</sup> MC self-burst microcapsules. The fact that sufficient control efficacy can be obtained without the added use of spreading agents has already been confirmed by publicly consigned tests by the Japan Plant Protection Association.

#### Safety

Pyriproxyfen, which is the active ingredient of Pluto<sup>®</sup> MC, is an analog of an insect growth regulator (IGR), and it is classified as a juvenoid insecticide. This

Table 10	Toxicological and ecotoxicological infor-
	mation of Pluto® MC

Animal toxicity			
Acute oral toxicity	Rat ♀	LD50 >2000 mg/	
Acute dermal toxicity	r Rat ♂♀	LD50 >2000 mg/kg	
Eye irritation	Rabbit	Minimally irritat	
Skin irritation	Rabbit	Mildly irritating	
Skin sensitization	Guinea pig	Not sensitizing	
Aquatic organism toxic	city		
Fish toxicity	Carp	LC50(96hr) 48 mg/	
Daphnia toxicity	Daphnia magna	a EC50(48hr) 31 mg/	
Toxicity to algae	Algae	EbC50(0-72h	r) 9.6 mg/L

compound works only on insects belonging to several taxonomic groups, and its effects on organisms in the environment outside that range are limited. Its effects on fish, alga and invertebrates are small,<sup>10)</sup> and its effects on mammals are also small.<sup>1</sup> It is also clear that the adverse effects of Pluto<sup>®</sup> MC on mammals and aquatic life are also extremely small (**Table 10**).

Furthermore, the adverse effects on beneficial insects that may occur during actual use in tea fields and the chemical damage to the tea were investigated as follows.

#### 1. Effects on silkworms

Even in very small amounts, it is known that pyriproxyfen, which is the active ingredient of Pluto<sup>®</sup> MC, inhibits cocoon formation in the silkworm.<sup>1)</sup> Sufficient steps must be taken to prevent accidents that damage silkworms. As a means of avoiding drift during spraying, we limited the period of use to the winter as described above and used the product in an MC formulation. Besides this, however, there is the danger that the active ingredient may be vaporized by heating from burning empty bottles of this formulation under inappropriate conditions. The fact that silkworm cocoon formation will actually be damaged by the active ingredients that have been vaporized by heating was confirmed by burning tests carried out on Lano® tape.2) If the containers for this insecticide are disposed of in conditions where incomplete burning easily arises, such as by burning in fields at low temperatures, there is a danger that the vaporized active ingredient will scatter in ways that are unforeseen.

Therefore, to prevent incidences of silkworm toxicity because of inappropriate handling of empty used bottles, we have implemented thorough sales and recovery management for this insecticide. Specifically, only tea farmers that are registered as members can purchase and use the insecticide during a specific period, and only by reservation. The empty used bottles are quickly recovered, and disposal of the containers is carried out by burning them in a high-performance, hightemperature furnace capable of completely burning and decomposing the active ingredient (at a temperature of 800°C inside the furnace). In addition, in the prefectures where the recovery system has been established according to the respective prefectural industrial waste treatment methods, sales commenced in stages which were limited to areas in locations where there was safe separation from locations with sericulture. Therefore, we were thorough in taking various measures to avoid incidences of silkworm toxicity not only in the technical aspects of this insecticide, but also in the distribution aspects.

#### 2. Effects on natural parasitic wasps

Branches with mulberry scale parasitism that had not been treated with insecticides and where the parasitism of natural parasitic wasps was foreseen were collected from tea fields in July, and these were immersion treated in various insecticides to check the inhibitory activity on adult emergence for natural parasitic wasps. As a result, about the same level of adult emergence in parasitic wasps was found for *Pteroptrix orientalis* and *Marietta carnesi* as in untreated areas. Because of this, we determined that the adverse effects on parasitic wasps were low (**Table 11**).

#### 3. Effects on Pseudoscymnus hareja

We examined the effects on *Pseudoscymnus hareja*, which is an important natural enemy of mulberry scale. Treated branches were collected in June from tea fields that had been treated with the insecticide in March, and mature larvae of *Pseudoscymnus hareja* were released onto these. After that, the pupation rate and the adult emergence rate were examined.

The results we observed were that the pupation rate for treated samples was the same as for untreated ones, but the adult emergence rate was 4.4%, so adult emergence inhibition was exhibited and an adverse impact was found (**Table 12**).

 Table 11
 Influence of pyriproxyfen (Pluto<sup>®</sup> MC) to the parasitic wasps emergence

Chemicals	Conc.	# of emerged	# of emerged wasps from female adult scale at 10 days after treatment				
	(ppm)	Pteroptrix orientalis	Marietta carnesi	Aphytis diaspidis	Total		
pyriproxyfen MC	90	23	11	0	34		
	900	17	12	3	32		
DMTP EC	400	0	0	0	0		
untreated		21	17	2	40		

	Shizuoka Pr	efecture Research Ins	titute of Agriculture of	& Forestry Tea Rese	arch Center (2005,	Kikukawa, Shizuoka)
Chemicals	Conc.	Application	Releasing*	# of test	Pupation	Emergence
Chemicais	(ppm)	date	date	old larvae	rate	rate
pyriproxyfen MC	90	9 <sup>th</sup> , May	12 <sup>th</sup> , June	49	91.8%	4.4%
DMTP EC	400	29 <sup>th</sup> , May	12 <sup>th</sup> , June	59	94.9%	83.9%
untreated		_	12 <sup>th</sup> , June	62	98.4%	86.9%

#### Table 12 Influence of pyriproxyfen (Pluto<sup>®</sup> MC) to predatory coccinellid (*Pseudoscymnus hareja*)

Shizuoka Prefecture Research Institute of Agriculture & Forestry Tea Research Center (2005, Kikukawa, Shizuoka)

\*: Full-grown larvae of P. hareja were released onto treated tea.

## 4. Effects on prevalence of occurrence of natural enemies in the field

We periodically examined the prevalence of occurrence of parasitic wasp adults, predatory midge adults and *Pseudoscymnus hareja* adults in fields where Pluto<sup>®</sup> MC had been used using yellow sticky traps hung on the plants (**Table 13**). Our results showed that the amount of *Arrhenophagus albitibiae* occurring during the first generation occurrence of scale insects was greater in the Pluto<sup>®</sup> treated area than in the areas with customary prevention. Occurrences of predatory midge adults, *Pseudoscymnus hareja* adults and spiders were substantially the same as in the customary areas throughout the test period. No clear adverse effect on any natural enemies was found.

A strong action on adult emergence inhibition was found for *Pseudoscymnus hareja* (Table 12), but this insect is a euryphagous natural enemy and is populated widely outside of tea fields, so its numbers are also affected by insects coming into the tea fields from outside, and on the field level, we presumed that there would be no clear adverse effects found on this insect. In addition, the amount of the various natural enemies occurring was not only affected by the formulation, but also depends on the amount of food and on the number of host insects, so we can assume that it is not possible to judge whether there has been an effect or not simply from the number.

Since we have not ascertained resurgence for the various harmful insects in the Pluto<sup>®</sup> MC treated areas at present, we presume that there is little adverse effect on natural enemies at the field level. We expect to clarify this issue in future research.

#### 5. Phytotoxicity to tea plants

We investigated phytotoxicity of Pluto<sup>®</sup> MC to the main varieties of tea plants. As a result, we found no phytotoxicity in the various varieties of yabukita (10 examples), yamanibuki (1 example), kanayamidori (1 example), komakage (1 example), okumidori (1 example) and asatauyu (1 example).

			Sumitomo Che	emical Co., Ltd. (2007, Ma	akinohara, Shizuoka )		
Natural enemies	Treatment block	# of catches insects / yellow sticky trap <sup>1</sup>					
Ivatural enemies	Treatment block	$1^{st} G^{\#}(4/27-7/7)$	2nd G(7/8-9/2)	3 <sup>rd</sup> G(9/3-10/29)	Total		
Parasitic wasps <sup>†</sup>	Pluto control *	347	173	29	549		
	Conventional control **	262	102	96	460		
Tricontarinia sp.	Pluto control	13	25	4	42		
Theomarinia sp.	Conventional control	12	24	14	50		
Pseudoscymnus hareja	Pluto control	16	2	0	18		
1 sendoscymnas nareja	Conventional control	14	6	1	21		
Spiders	Pluto control	9	17	6	32		
Spider 5	Conventional control	11	5	7	23		

 Table 13
 Seasonal prevalence of occurrence of natural enemies on mulberry scale in the tea field applied with pyriproxyfen (Pluto<sup>®</sup> MC)

\* : Pyryproxyfen MC (90ppm, 1000L/10a) was applied on 3rd, May.

\*\* : Buprofezin and fenproxymate FL (200ppm and 40ppm, 1000L/10a) was applied on 21<sup>st</sup>, May and DMTP EC (400ppm, 1000L/10a) was applied on 23<sup>rd</sup>, July.

 $\int$ : Trap size : 10cm × 10cm. (2 reps)

# : G; generation in a year.

† : The dominant species was Arrhenophagus albitibiae.

Target crops	Target pests	Dilution rate	Spray volume	Application timing	Times*
				Winter dormant period of female adult	
Tea	Mulberry scale	fx1000	1000L/10a	(Until 45 days before first pluck	1
				and before tea germination)	

#### Table 14 Domestic registration of Pluto<sup>®</sup> MC

\* : Maximum application times in any given season

#### Conclusion

In recent years, there has been a trend towards an expansion in the problem of mulberry scale in tea cultivation in Japan, and tea farmers, who are aging, are being spurred on by the escalating problem of control requiring a large amount of time and effort. To introduce pyriproxyfen, which has a superior control efficacy on scale insects, while making every effort to avoid adverse effects on the silkworm industry, we developed Pluto<sup>®</sup> MC as a laborsaving product that can be used on-site safely and with confidence to greatly reduce the labor as a result of earnest investigations in areas such as basic study and special formulation technology. With the support and guidance of the various supervisory organizations involved, we finally obtained agricultural chemical registration (Table 14) in December 2007 and brought commercialization to fruition.

There are cases where control of mulberry scale has used a maximum of six components with three applications of insecticides per year depending on the area for control, but it is possible to greatly reduce the number of components and the number of times for using agricultural chemicals to one component once per year by using this insecticide. Not only is the required labor reduced, but also we can expect it to contribute to savings in fuel costs for sprayers, which have risen steeply in recent years. In addition, we can assume that by using this insecticide which has a high level of safety for both humans and animals in the winter before the sprouting of the first tea sprouts, we are responding to the increasing needs of general consumers for safety and confidence. Going forward, we hope that Pluto<sup>®</sup> MC will be used widely at tea production sites as an agricultural resource that is laborsaving and has high performance and a high level of safety, and that it is a landmark technology that responds to the demands of producers and general consumers.

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