

The effect of sawdust fermentation period and storage period after sawdust fermentation on the development of *Prottaetia brevitarsis* larvae

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Abstract

To investigate the effect of sawdust fermentation period and storage period after fermentation on development as food for *P. brevitarsis* larvae, individual and group breeding were conducted on berry sawdust and oak sawdust. In individual breeding, the growth period of *P. brevitarsis* larvae was reduced by 12 days from the 60-day fermentation of berry sawdust to the 40-day fermentation of berry sawdust, 30 days from the 90-day fermentation of oak sawdust, and the weight of the larvae was the heaviest. In group breeding, the time it takes for *P. brevitarsis* larvae to change from 1st to 3rd instar is about 30 days after hatching from 60-day fermentation of berry sawdust, while 90-day fermentation of oak sawdust took more than 60 days, so the growth speed was fast and the survival rate was good. The results of the farmhouse demonstration test were the same trend, and it was judged that it would be possible to produce *P. brevitarsis* larvae with berries fermented sawdust, and it would be advantageous in terms of economy. In addition, for both individual and group breeding, the growth period of *P. brevitarsis* larvae was longer as the storage period was longer, the weight of the larvae decreased, and the survival rate was no different. The development period of *P. brevitarsis* larvae was the longest in the storage period of 18 months for berries fermented sawdust, and the storage period of oak fermented sawdust was longer in the storage period of 12 months and 18 months. Therefore, considering the results of individual breeding and group breeding, the fermentation period was appropriate for 60 days for berries sawdust, and the storage period for berries sawdust was stable from 0 to 12 months, and 0 to 6 months for oak sawdust.

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Introduction

In 2020, the production industry was the largest in Korea (98.5%), and the production type of industrial insects was 66.5% for edible and medicinal, 12.8% for feed, and 20.7% for pets, and 69.6% of *P. brevitarsis* was produced among edible and medicinal

insects (Kim *et al.*, 2022; Ban *et al.*, 2022; Choi *et al.*, 2021).

Prottaetia brevitarsis is an insect belongs to Cetoniidae, Coleoptera that feeds on the woody parts of plants. In particular, the *P. brevitarsis* larvae cause damage the roots of the plant's underground, and use the cellulose of wood as a nutritional source. However, it is said that insect microorganisms or

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symbiotic bacteria living in the posterior intestines mainly induce chemical cellulose decomposition because the *P. brevitarsis* larvae does not produce enzymes that decompose cellulose on its own (Bayon and Mathelin, 1980, Lemke *et al.*, 2003; Lo *et al.*, 2003; Kang *et al.*, 2005).

Oak sawdust is used as a food source for breeding *P. brevitarsis*, but the use of oak sawdust inevitably causes damage to oak trees, must be fermented for a certain period of time to be used as *P. brevitarsis* larvae feed, and usually requires a long period of 60 days or more.

In addition, most of the remaining sawdust used as *P. brevitarsis* feed after the fermentation of sawdust is considered to have lost its value as larval feed, and the feed cost for rearing 3.3 m² is 106,000 won, accounting for 32% of the total production cost (Lee *et al.*, 2018). Therefore, it is essential to develop low-cost fermented sawdust and insect productivity improvement technology to reduce management costs.

On the other hand, pruning trees of about 60,000 ha of Korean internal fruit trees such as apples, pears, and peaches, and an abandoned trees of mushroom cultivation are considerably occurring. In particular, in the case of orchards, waste trees that are discarded or burned to be reproduced as new species and vestibule trees that are generated through pruning every year are generated, so recycling is necessary from an industrial perspective.

Even from an environmental point of view, research has been conducted to recycle waste trees into *P. brevitarsis* feed for the purpose of shortening the period during which they are decomposed by microorganisms and circulated to nature in terms of removing harmful factors to the natural ecosystem (Kang *et al.*, 2005).

Berry crops such as mulberry, raspberry, and blueberry have a cultivation area of 2,994 ha in 2020, and pruning trees are generated every year, and berry sawdust that fermented berry pruning trees is reported to be advantageous in breeding *P. brevitarsis* compared to oak sawdust (Lim *et al.*, 2022b).

In addition, research on alternative feed for *P. brevitarsis* has been conducted in various ways, such as after mushroom harvesting culture medium, soybean-curd residues, and mulberry trees, and has been reported to be effective in the development of *P. brevitarsis* larvae (Lim *et al.*, 2022a, b, c). But the fermentation methods, fermentation process, and storage methods have not been dealt with. In particular, since alternative feed has different ingredients from oak sawdust, it is thought that a careful review

of the fermentation method is needed.

Therefore, this study was conducted to provide basic data on the fermentation method of sawdust by comparing and analyzing the effect of the fermentation period of oak sawdust and berries sawdust and the storage period after sawdust fermentation. In addition, demonstration tests were conducted at insect breeding farms to recognize the value and importance of *P. brevitarsis* feed.

Materials and methods

Experimental insects and sawdust

In April 2021, adults of *P. brevitarsis* were purchased from breeding farms in Jangsu-gun, Jeollabuk-do, and that were raised put in a 20 L plastic box (543 × 363 × 188 mm)/120 adults (female: male = 2: 1) of *P. brevitarsis* containing berries mixed fermented sawdust at a breeding room (25±1°C, RH 50-60%) in the Sericulture and Entomology Experiment Station, Jeollabuk-do Agricultural Research and Extension Services. And it was take eggs, and it was hatched larva used for experiments.

As for adult prey of *P. brevitarsis*, banana fruits sold on the market were purchased and replaced every two to three days to be supplied, so that they could be take eggs every week until the adult life was over. The takes eggs were stored separately to induce hatching, and the experiment was conducted by controlling the density of the hatched larvae.

The berries sawdust used in the experiment was collected pruning branch from 2020 for a year from mulberry, raspberry, and blueberry cultivated in Jeollabuk-do, mix crushed using a twig shredder (DY13-838, Dongyang Industrial) and shredder (MC10SS5-F, Seongchang Machine), and used a 4 mm stainless steel net to make sawdust and store it in a 20 kg bag. Oak sawdust was purchased and used at breeding farm of *P. brevitarsis* in Iksan.

The effect of sawdust fermentation period

1. Individual breeding

The previously prepared berries sawdust and oak sawdust were mixed with ingredients, respectively, and fermented from April to June 2021 at the fermentation room of breeding farm of *P. brevitarsis* in Iksan City with different fermentation periods.

During the fermentation period of sawdust, berries sawdust was fermented for 40 days and 60 days, respectively, and

oak sawdust was fermented for 90 days, and supplied as food for *P. brevitarsis* larvae, investigating the larva development characteristics according to the sawdust fermentation period.

For larval breeding, petri dish ($\phi 100 \times 40$ mm) was filled with fermented sawdust with different fermentation periods, and then one larva, the first day of hatching, was put in and raised individually. Thirty of *P. brevitarsis* larvae were investigated for each treatment, and repeated three times.

As for the growth characteristics of *P. brevitarsis* larvae, the growth period by age, weight, and mortality were investigated by sawdust fermentation period. The larval growth period and average weight of the larvae were investigated once every two to three days, and the mortality rate was investigated by checking whether they survived. The larval stage was classified by measuring the size of the larval head.

2. Collective breeding

In order to compare it with individual breeding, a group breeding of *P. brevitarsis* larvae was conducted, and demonstration tests were conducted at two *P. brevitarsis* breeding farms in Iksan and Jangsu-gun compared to the Sericulture and Entomology Experiment Station, and the larva development characteristics were investigated according to the fermentation period.

The fermentation period, like individual breeding, berries sawdust was fermented on 40 days and 60 days, and oak sawdust was fermented for 90 days.

For group breeding, a 20 L living box (plastic box; 543 mm \times 363 mm \times 188 mm) was used, and after filling about two-thirds of fermented sawdust with different fermentation periods, respectively, 100 freshly hatched larvae were put in per living box and repeated three times.

The larval growth characteristics of each fermentation period according to group breeding were investigated from the 30th day after the start of breeding to the 100th day at intervals of 10 days, and the mortality rate was converted by checking the survival.

In the demonstration test, the quantity of *P. brevitarsis* larvae was compared the oak sawdust fermented on the 90th. And the berries sawdust fermented on the 40th was raised at farms in Jangsu-gun, and the berries sawdust fermented on the 60th was raised at farms in Iksan city. 180 larvae on the 14th day after hatching per living box were raised, respectively, and were raised in 12 living boxes for each treatment.

As for the larval quantity, the survival rate by treatment and the

weight per living box were compared, and the average weight of the individual was converted and compared.

Effect of fermented sawdust storage period

1. Individual breeding

To investigate the storage properties of remaining fermented sawdust after sawdust fermentation, stored sawdust at storage warehouse (constant temperature) for 0, 6, 12, and 18 months after sawdust fermentation was supplied as food for larvae, and each development characteristic was investigated.

Berries sawdust and oak sawdust were compared, and stored berries sawdust was fermented for 60 days, and stored oak sawdust was fermented for 90 days.

For the breeding of *P. brevitarsis* larvae, petri dish ($\phi 100 \times 40$ mm) was filled with fermented sawdust with different storage periods, and then one larva, the first day of hatching, was added and raised individually. Thirty of *P. brevitarsis* larvae were investigated for each treatment, and repeated three times.

As for the development characteristics, the development period and weight of *P. brevitarsis* larvae by age were investigated by storage period, and the survival rate and weight were investigated during the harvest period.

The growth period and weight of each larval age were investigated once every two to three days, the larval age was classified by measuring the size of the larval head, and the larval weight was compared with the average and maximum weight of each individual.

The survival rate and average weight of the larval harvest season were converted by examining the survival number just before pupa and the weight of each individual.

2. Collective breeding

In order to compare individual breeding and group breeding, group breeding was conducted using sawdust used for each storage period after the fermentation of sawdust in the previous individual breeding, and the development characteristics of *P. brevitarsis* larvae were investigated according to the storage period of fermented sawdust.

For group breeding, a 20 L living box (plastic box; 543 mm \times 363 mm \times 188 mm) was used, and after filling about two-thirds of fermented sawdust with different storage periods, 100 freshly hatched larvae were added per living box and repeated three times.

The growth characteristics of *P. brevitarsis* larvae according to group breeding were investigated for the survival rate and

Table 1. Developmental characteristics of *P. brevitarsis* larvae on different fermented periods of berries sawdust and oak sawdust in individual breeding

Fermentation periods (day)	Developmental period (day)				Larva weight (g)			Mortality rate (%)	
	1 st	2 nd	3 rd	Total	1 st	2 nd	3 rd		
Berries sawdust	40	15.5 b	24.2 b	55.4 ab	95.1 b	0.06 b	0.40 b	2.4 b	1.0 a
	60	10.7 a	18.5 a	54.0 a	83.2 a	0.07 a	0.53 a	2.5 a	0.0 a
Oak sawdust	90	19.4 c	28.1 c	66.0 c	113.5 c	0.05 c	0.31 c	1.9 c	2.7 a

* Different letters in a column indicate different significance by the Duncan's multiple range test.

weight of larvae during the harvest period, and the survival rate and weight of larvae per treatment and living box were converted into an average.

Statistical processing

The results obtained from the experiment were one-way ANOVA using the PASW Statistics 18 (SPSS Inc., Chicago, US) program, and then T-test and Duncan multiple tests were conducted at a 5% significance level to test the difference between treatment means.

Results and Discussion

The Effect of Sawdust Fermentation Period on the Development of *P. brevitarsis* larva

1. Individual breeding

According to the results of the survey on the growth characteristics of *P. brevitarsis* larvae (Table 1) in individual breeding, the total growth period of berries sawdust by 60-day fermentation to 83.2 days was shortened by 12 days from 95.1 days of 40-day fermentation. Compared to 113.5 days of oak sawdust by 90-day fermentation, it was shortened by 30 days, and a significant difference was recognized.

The development period for each larval instar at berries sawdust by 60-day fermentation was also reduced by 4.8 days for 1st instar and 5.7 days for 2nd instar, and there was no difference in 3rd instar, compared to 40 days of fermentation. And compared to oak sawdust by 90-day fermentation, it was shortened by 8.7 days for 1st instar, 9.6 days for 2nd instar, and 12 days for 3rd instar, and significant difference was recognized.

In addition, the average weight by larval instar of berry sawdust by 60-day fermentation was 0.07 g for 1st instar, 0.53 g for 2nd instar, and 2.5g for 3rd instar, up from 40-day fermentation

and 90-day fermentation of oak sawdust, and there was no significant difference in mortality.

This shows that the development period of 60-day fermentation of berry sawdust is shorter and heavier than the 40-day fermentation of berries sawdust and 90-day fermentation of oak sawdust, and the 60-day fermentation period of berry sawdust is more appropriate than the 40-day fermentation period, which is too short fermentation period.

In addition, it was similar to the report (Lim *et al.*, 2022b) that the growth period of larvae in berry sawdust is shorter than that of oak sawdust, and it is considered economically advantageous.

On the other hand, depending on the type of microorganism in fermented sawdust, it is judged that changes in microbial phase over the fermentation period affect the feeding activity of *P. brevitarsis* larvae, such as reports that do affect or not affect shiitake mycelium, growth, and growth promotion (Koo *et al.*, 2014; Han, 2003; Shim *et al.*, 2002). But further review is expected.

2. Collective breeding

As a result of examining the growth rate according to the fermentation period of sawdust in the group breeding of *P. brevitarsis* larvae using berries sawdust (Table 2), It took 30 days from 60-day sawdust fermentation to 3rd instar after hatching, 40 days from 40-day sawdust fermentation, and 60 days after hatching in oak sawdust.

During the development period, the mortality rate was the lowest from 60-day fermentation to 3.0%, compared to 5.7% on 40-day fermentation and 14.0% on 90- day fermentation of oak sawdust. In addition, in group breeding, the mortality rate was higher than that of individual breeding (Table 1), so it is believed that there was competition among individuals.

According to the number of deaths of *P. brevitarsis* larvae, they appear to die within 30 days of early breeding, and the

Table 2. The number and mortality of larva by instar of *P. brevitarsis* according to the fermentation period of sawdust from berries and oak sawdust in group breeding

Fermentation periods (day)		No. of Larvae on developmental periods								Mortality rate (%)	
		30 day	40 day	50 day	60 day	70 day	80 day	90 day	100 day		
Berries sawdust	40	1 st	0	0	0	0	0	0	0	0	5.7 ab
		2 nd	122	3	2	2	1	1	1	0	
		3 rd	168	284	283	268	241	225	127	79	
	60	1 st	0	0	0	0	0	0	0	0	3.0 a
		2 nd	3	0	0	0	0	0	0	0	
		3 rd	251	105	29	13	9	6	0	0	
Oak sawdust	90	1 st	0	0	0	0	0	0	0	0	14.0 b
		2 nd	276	105	53	9	6	3	2	0	
		3 rd	0	170	219	261	259	259	259	259	

* Different letters in a column indicate different significance by the Duncan's multiple range test.

Table 3. Productivity of *P. brevitarsis* on different fermentation periods of berries sawdust and oak sawdust in a demonstration test place of Jangsu and Iksan (collective breeding)

fermentation period (day)	Breeding place	Quantities of <i>P. brevitarsis</i> larva				
		Survival rate (%)	Larval weights (g/box)	Larval weights (g/individual)	Breeding period (day)	
Berries	40	Jangsu	92.2 a	382.6 ab	2.1 b	48
sawdust	60	Iksan	88.8 ab	409.0 a	2.5 a	60
Oak sawdust	90	Jangsu	82.4 b	303.0 b	1.9 b	48
	90	Iksan	87.8 ab	388.0 ab	2.5 a	60

* Different letters in a column indicate different significance by the Duncan's multiple range test.

growth of larvae in berries sawdust is relatively stable compared to oak sawdust that dies steadily until the end of breeding. It is believed that it is necessary to review the nutrients or harmful substances contained in sawdust.

Therefore, considering the rate of development and mortality in group breeding, it is estimated that the rate of development is fast, production is the highest at 60-day fermentation, even if the fermentation period of sawdust is too short or long, it is judged to be disadvantageous for the development of *P. brevitarsis* larvae.

As a result of investigating the effect of sawdust fermentation period on the quantity of *P. brevitarsis* larvae in the demonstration test (Table 3), according to the fermentation period the survival rate did not differ in the 40th and 60th fermentation of berries sawdust, but a significant difference was recognized in the 90th fermentation of oak sawdust in Jangsu.

The weight per living box was the highest at 409 g/box in the 60-day fermentation of berries sawdust in Iksan, but there was no difference from the 40-day fermentation of Jangsu, and like the survival rate, a significant difference was recognized as 303 g/box in the 90-day fermentation of oak sawdust in Jangsu. However, the 90-day fermentation of oak sawdust in Iksan was 388 g/box, and no significant difference was recognized from berries sawdust.

In addition, as a result of converting the weight of each individual using the survival rate and weight per living box, the 60-day fermentation of berries sawdust and 90-day fermentation of oak sawdust in Iksan were heavy at 2.5 g, while the 40-day fermentation of berries sawdust in Iksan and 90-day fermentation of oak sawdust in Jangsu were the same results, with significant differences in weight of 2.1 g and 1.9 g, respectively.

This seems to be due to the fact that Iksan farmers harvested it in 60 days, and Jangsu farmers harvested it in 48 days after breeding, and it is judged that a period of sufficient development of *P. brevitarsis* is needed.

However, in Jangsu, the survival rate of 40-day fermentation of berries sawdust was expected to increase by more than 10% compared to the 90-day fermentation of oak sawdust, and the weight of larvae was significantly higher, so the quantity was expected to increase. But in Iksan, there is no difference in the survival rate and larval weight between 60-day fermentation of berries sawdust and 90-day fermentation of oak sawdust, so it is believed that *P. brevitarsis* can be produced with berries sawdust. It is judged to be advantageous in terms of economy, but it is necessary to comprehensively consider the results of individual and group breeding in the future, and further review is needed.

In addition, as a result of a survey on the current status of leading edible insect breeding farms in 2017, the larval period and the weight of last larvae differed between farmers. And as shown in the report (Song *et al.*, 2017), each farm adds various ingredients to oak sawdust as a source of food to increase productivity of *P. brevitarsis*, it is necessary to standardize of *P. brevitarsis* breeding technology in actual farms.

Considering the individual and group breeding of *P. brevitarsis* larvae according to the fermentation period of berries sawdust, and the results of a demonstration tests, the fermentation period of berries sawdust is considered appropriate for 60 days. Apart from the fermentation period in individual and group breeding, it is judged that berries sawdust develops faster than oak sawdust.

This is similar to a report that the development period of *P. brevitarsis* larvae in berries fermented sawdust was short and the weight increased (Lim *et al.*, 2022b). And according to the report (Kim and Kang, 2005), the development period is shorter than that of under fermented sawdust, and is significantly shorter than in 1st to 2nd instar of *P. brevitarsis* larvae. It is consistent with the view that berries sawdust is fermented relatively quickly compared to oak sawdust.

Effect of Fermented Sawdust storage period

1. Individual breeding

According to the results of the survey on the development characteristics of *P. brevitarsis* larvae according to the storage period of fermented sawdust in individual breeding (Table 4). The total development period of *P. brevitarsis* larvae in berries

fermented sawdust was the longest from 18 months of storage period to 164.8 days, and there was no difference between 154 and 156 days for 0-12 months. Oak fermented sawdust did not differ from 173 to 176 days until 6 months of storage period, and 12 months and 18 months of storage period had a long development period of 182.3 days and 184.7 days, respectively, so a significant difference was recognized.

The development period by larval instar of *P. brevitarsis* was the shortest from 0 months of storage period to 12.9 days for 1st instar in berries fermented sawdust, and the longest from 18 months of storage period to 14.1 days. There was no difference in 2nd instar, and 3rd instar was the longest from 18 months of storage period to 130.2 days, and a significant difference was recognized. Oak fermented sawdust had a long development period of 12-18 months of storage period in all 1st to 3rd instar.

In addition, oak fermented sawdust had a longer development period than berries fermented sawdust, and 1st instar of *P. brevitarsis* was 2-7 days, 2nd instar was 4-6 days, and 3rd instar was 18-25 days longer.

In individual breeding, the average weight of larvae according to the storage period of berries fermented sawdust was not much different in 1st and 2nd instar of *P. brevitarsis* larvae from 0 to 12 months of storage period, but 18 months of storage period reduced in weight, indicating a significant difference in 3rd instar.

There was no difference at 1st and 2nd instar of *P. brevitarsis* larvae in the maximum weight of larvae, but 3rd instar was the heaviest at 0 months of storage period, and there was no difference between 6 and 18 months of storage period in berries fermented sawdust. There was no difference in the average weight and maximum weight of 1st and 2nd instar *P. brevitarsis* larvae by storage period of fermented sawdust in oak fermented sawdust, but the weight of 3rd instar decreased from 12 months and 18 months of storage period.

Therefore, in terms of the growth period and the weight of larvae in individual breeding, the storage period of fermented sawdust affects the development of *P. brevitarsis* larvae, and it is expected to affect the growth of berries fermented sawdust for 18 months and oak fermented sawdust for more than 12 months of storage period.

In harvest time of *P. brevitarsis* larvae by individual breeding used fermented sawdust, as a result of the survey on the growth characteristics of *P. brevitarsis* larvae according to the storage period of fermented sawdust (Table 5), there was no difference in the storage period of berries fermented sawdust with a survival

Table 4. Developmental periods and weights of *P. brevitarsis* larvae according to the storage period of fermented sawdust in individual breeding

Storage period (Month)	Developmental period (days)				Weights of larvae (g)						
	1 st	2 nd	3 rd	Total	Average			Max			
					1 st	2 nd	3 rd	1 st	2 nd	3 rd	
Berries fermented sawdust	0	12.9a	20.7a	120.8a	154.4a	0.08a	0.57a	2.54a	0.10a	0.67a	3.01a
	6	13.8a	21.1a	120.1a	156.0a	0.06a	0.58a	2.42ab	0.09a	0.59a	2.70b
	12	13.6a	20.6a	122.3a	156.5a	0.06a	0.60a	2.40ab	0.10a	0.64a	2.79b
	18	14.1a	20.5a	130.2b	164.8b	0.07a	0.62a	2.29b	0.09a	0.64a	2.73b
Oak fermented sawdust	0	18.4ab	21.9ab	131.0a	173.3a	0.06a	0.40a	1.90ab	0.08a	0.50a	2.59a
	6	16.4a	21.0a	138.7bc	176.1ab	0.06a	0.41a	2.01a	0.08a	0.51a	2.48ab
	12	20.4b	26.9b	135.0b	182.3b	0.05a	0.39a	1.76b	0.08a	0.51a	2.23c
	18	20.3b	25.9b	140.5c	184.7b	0.06a	0.41a	1.75b	0.08a	0.51a	2.37b

* Different letters in a column indicate different significance by the Duncan's multiple range test.

Table 5. Survival rate and weight of *P. brevitarsis* larva according to the storage period of fermented sawdust in individual breeding

Storage period (Month)	Berries fermented sawdust		Oak fermented sawdust	
	Survival rate (%)	Weights of larvae(g)	Survival rate (%)	Weights of larvae(g)
0	99.9 a	2.6 a	100 a	2.4 a
6	99.7 a	2.6 a	100 a	2.4 a
12	99.7 a	2.5 a	100 a	2.0 b
18	99.8 a	2.5 a	99.9 a	1.9 b

* Different letters in a column indicate different significance by the Duncan's multiple range test.

rate of more than 99%, and the weight of larvae was also 2.5 to 2.6 g, and no significant difference was recognized according to the storage period.

All storage periods of oak fermented sawdust had a survival rate of almost 100%, and there was no significant difference. Like berries sawdust, the weight of *P. brevitarsis* larvae was similar from 0 months and 6 months of storage period to 2.4 g, but decreased to 2.0 g and 1.9 g, respectively, in 12 months and 18 months, and significant differences were recognized.

Therefore, considering the growth period and larval weight *P. brevitarsis* larvae according to the storage period of fermented sawdust in the previous individual breeding, in berries fermented sawdust, there is no difference in the survival rate of *P. brevitarsis* larvae according to the storage period in the individual breeding, but it is judged relatively stable until 12 months of storage period in terms of the weight of the larvae. Oak fermented sawdust is believed to reduce the weight of larvae

if the storage period is more than 12 months, which interferes with the development of larvae.

2. Collective breeding

Results of survey the survival rate and weight of *P. brevitarsis* larvae according to the storage period of fermented sawdust during the harvest season in group breeding (Table 6), berries fermented sawdust had the lowest survival rate of 87.3% in the storage period of 18 months, and the weight of larvae was also the smallest at 231.6 g per 100 animals, and a significant difference was recognized.

Oak fermented sawdust had the lowest survival rate of 86.3% in 0 months of storage period, but there was no significant difference, and on the contrary, the larvae weight was the lowest in 18 months of storage period, like berries fermented sawdust, and significance was recognized.

Therefore, as with the results of individual breeding, berries

Table 6. Survival rate and weight of *P. brevitarsis* larva according to the storage period of fermented sawdust in group breeding

Storage period (Month)	Berries fermented sawdust		Oak fermented sawdust	
	Survival rate (%)	Weights of larva (g/Living box)	Survival rate (%)	Weights of larva (g/Living box)
0	93.0 ab	240.2 ab	86.3 a	196.2 a
6	92.0 ab	239.3 ab	91.0 a	198.3 a
12	96.0 a	243.7 a	89.7 a	195.7 a
18	87.3 b	231.6 b	93.3 a	189.6 b

* Different letters in a column indicate different significance by the Duncan's multiple range test.

fermented sawdust seems to interfere with the development of *P. brevitarsis* larvae at a storage period of 18 months in group breeding, oak fermented sawdust has no difference in survival rate compared to individual breeding, and the weight of larvae is believed to affect the growth of larvae from 12 months' storage period in individual breeding and 18 months of group breeding.

Considering the results of individual and group breeding of *P. brevitarsis* larvae, there is no difference in survival rate as the storage period of fermented sawdust increases, but it affects the growth period and the weight of the larvae. Berries fermented sawdust is considered stable up to 12 months of sawdust storage period because it interferes with the development of larvae during the storage period of 18 months. Oak fermented sawdust is believed to affect the development of larvae from 12 months of sawdust storage in individual breeding and group breeding was from 18 months of sawdust storage period, so it was judged that more than the storage period of 12 months would interfere with the development of larvae.

In the above, considering the growth period, larval weight, and survival rate of *P. brevitarsis* larvae according to the fermentation period of sawdust and storage period after fermentation, the fermentation period of berries sawdust is appropriate for 60 days, and the storage period is appropriate for 0 to 12 months. It is believed that the storage period after fermentation of oak sawdust, which is previously used as food, is appropriate for 0 to 6 months.

In addition, because the fermentation period of berries sawdust is shorter, the growth period of *P. brevitarsis* larvae is shorter, and the survival rate and larval weight are increased compared to oak sawdust, it is estimated that berries sawdust will be more effective in raising *P. brevitarsis* larvae in economic terms.

References

- Bayon C, Mathelin J (1980) Carbohydrate fermentation and by-product absorption studied with labelled cellulose in *Oryctes nasicornis* larvae (Coleoptera: Scarabaeidae). *J Insect Physiol* 26, 819-828. [https://doi.org/10.1016/0022-1910\(80\)90100-6](https://doi.org/10.1016/0022-1910(80)90100-6)
- Ban E, Kim BS, Choi R, Kim I, Seo M, Hwang JS, et al. (2022) Immune-enhancing effects of *Protaetia brevitarsis seulensis* larvae extracts on RAW264.7 macrophages. *Int J Indust Entomol* 45, 108-114. <https://doi.org/10.7852/ijie.2022.45.2.108>
- Choi I, Choi S, Son J, Jang W, Chung T (2021) Antioxidant and cytoprotective effects of enzyme extracted constituents of *Protaetia brevitarsis seulensis* powder. *Entomol Res* 51, 90-94. <https://doi.org/10.1111/1748-5967.12484>
- Han BN (2003) Microbe profile on the fermentation process for the diet of *Protaetia brevitarsis*. p. 63. MS Thesis, Chungnam national university, Daejeon.
- Kang SJ, Park CW, Han SC, Yi YK, Kim YG (2005) A grub (*Protaetia brevitarsis seulensis*) rearing technique using cellulose-digesting bacteria and natural recycling of rearing byproduct to an organic fertilizer. *Korean J Appl Entomol* 44, 189-197.
- Kim, HG, Kang KH (2005) Bionomical characteristic of *Protaetia brevitarsis*. *Korean J Entomol* 44, 139-144.
- Kim WK, Kim SY, Ji SM, Chang GD, Song JH (2022) Current status and future perspective of industrial insects use in South Korea. *Korean J Appl Entomol* 61, 221-227. <https://doi.org/10.1016/j.jpbi.2021.102139>
- Koo CD, Lee SJ, Lee HY, Park YW, Lee HS, Kim JS (2014) Changes on physio-chemical properties of oak sawdust during fermentation. *J Mushrooms* 12, 209-215. <https://doi.org/10.14480/JM.2014.12.3.209>

- Lee SB, Kim JW, Bae SM, Hwang YH, Lee BJ, Hong, KP, *et al.* (2018)
Evaluation of spent mushroom substrates as food for white-spotted
flower chafer, *Protaetia brevitarsis seulensis*(Coleoptera: Cetoniidae).
Korean J Appl Entomol 57, 97-104. [https://doi.org/10.5656/
KSAE.2018.04.0.011](https://doi.org/10.5656/KSAE.2018.04.0.011)
- Lemke T, Stingl U, Egert M, Friedrich MW, Brune A (2003)
Physicochemical conditions and microbial activities in the highly
alkaline gut of the humus-feeding larva of *Pachnoda ephippiata*
(Coleoptera: Scarabaeidae). Appl Environ Microbiol 69, 6650-6658.
<http://doi.org/10.1128/AEM.69.11.6650-6658.2003>
- Lim JR, Moon HC, Park NY, Lee SS, Kim W, Choi CH, *et al.* (2022a)
Optimal Larval Density and Low Temperature Storage Conditions
for Rearing of *Protaetia brevitarsis* (Coleoptera: Cetoniidae) using a
Fermented Mulberry Sawdust-base Diet. Korean J Appl Entomol 61,
555-562. <https://doi.org/10.5656/KSAE.2022.08.0.031>
- Lim JR, Moon HC, Park NY, Lee SS, Lee EJ, Nam JH, *et al.* (2022b)
Development and oviposition characteristics of *Protaetia brevitarsis*
(Coleoptera: Cetoniidae) fed with fermented sawdust from different
berries. Korean J Appl Entomol 61, 377-385. [https://doi.org/10.5656/
KSAE.2018.11.0.051](https://doi.org/10.5656/KSAE.2018.11.0.051)
- Lim JR, Moon HC, Park NY, Lee SS, Nam JH, Kim W, *et al.* (2022c).
Development and oviposition characteristics of *Protaetia brevitarsis*
(Coleoptera: Cetoniidae) by additional feeding Korean black
raspberry marc and blueberry marc. Int J Indust Entomol 44, 44-54.
<https://doi.org/10.7852/ijie.2022.44.2.44>
- Lo N, Watanabe H, Sugimura M (2003) Evidence for the presence of a
cellulase gene in the last common ancestor of bilaterian animals. Proc
R Soc Lond B 270: 69-72. <http://doi.org/10.1098/rsbl.2003.0016>
- Shim MS, Kwon HG, Kim JH (2002) Composting of sawdust substrate
for growing Shiitake mushroom (*Lentinus edodes*). pp 143. The
Ministry of Agriculture and Forestry, Nati R&D Res Rep, Sejong.
- Song MH, Han MH, Lee SH, Kim ES, Park KH, Lim WT, *et al.* (2017).
A field survey on edible insect farms in Korea. J. Life Sci 27, 702-
707. <https://doi.org/10.5352/JLS.2017.27.6.702>