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Residual Effect of Chemical Insecticides used Against Thrips on Larval Parameters of Silkworm *Bombyx mori* L.

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ABSTRACT: A study was conducted to evaluate the safety period and residual effect of chemical insecticides used against thrips through rearing performance of silkworm *Bombyx mori* L. Insecticides were sprayed on mulberry at an interval of 25 (20 DAS), 30 (15 DAS) and 35 (10 DAS) days after pruning. Among the treatments acetamiprid recorded highest larval mortality per cent even upto 20 DAS, Mortality was zero in buprofezin, fipronil, dimethoate at 15 and 20 DAS. Where as in acephate and imidacloprid there was zero mortality at 20 DAS. Acetamiprid adversely affected the rearing performance of silkworm with residual toxicity symptoms like vomiting gut juice, rectal protrusion, stiffening of larval body and twisting of larval body in agony, hook shaped larvae with shrunken body, death of larvae without moulting, incomplete moulting and death of ripened larvae without spinning.

Keywords: Mulberry, silkworm, DAS, insecticides, acetamiprid.

INTRODUCTION

Since ancient times, silk has demonstrated its supremacy as a textile fiber, and no other fabric can match its shine and elegance till today. Mulberry *Morus* spp. (Urticales: Moraceae) is the sole food for silkworm, *Bombyx mori* L. However luxuriant growth of mulberry plants invites infestation of varied types of insect pest species resulting in considerable reduction in quantity and quality of leaf which adversely effects quantum of silkworm rearing and cocoon productivity. Thrips (*Pseudodendrothrips mori*), a serious sap sucker that was formerly known as *Belothrips mori*, is the most common sucking pest of mulberry, with an incidence of 42.55% (Anon., 1997). Among different species of the thrips infesting mulberry in Karnataka, *P. mori* is predominant in central, eastern and southern dry zone (Devaiah and Kotikal 1983). So far, 35 species of thrips are reported to inflict loss of yield to mulberry leaves all over the world, of which 21 species are from India. Nearly 40-50 per cent of leaf loss out of total leaf production is estimated due to this pest (Narayanaswamy *et al*., 1996). In the traditional areas, since silkworm rearing is practiced 4-5 times in a year, leaf production has to be achieved at a faster rate maintaining both the mulberry yield and quality characters. Farmers must maintain pest-free mulberry gardens in order to meet their hectic rearing schedules, which makes the cultural, biological and physical approaches time-consuming and impractical. In order to complete silkworm raising on schedule, insecticide spraying is therefore necessary to protect the plants against pests. Silkworm *B. mori* L. being an insect is also sensitive to insecticides, feeding them mulberry leaves treated with pesticides might result in different kinds of damage to the worms, depending on the type and concentration of pesticides employed. Safety period of the insecticides must be followed which ranges from 3 to 110 days depending upon the insecticide used (Kuribayashi, 1988).

MATERIALS AND METHOD

A. Experimental details

The details of field experiments conducted are given below.

Crop : Mulberry
Variety : Victory-1 (V-1)
Spacing : (90 + 150) × 60 cm (paired row)
No. of treatment : 7
No. of replication : 3
Design : RCBD

B. Treatment details

Treatments	Details	Dosage
T_1	Buprofezin 25 SC	2 ml/l
T_2	Acephate 75 SP	1 g/l
T_3	Acetamiprid 20 SP	0.2 g/l
T_4	Imidacloprid 17.8 SL	0.3 ml/l
T_5	Fipronil 5 SC	2 ml/l
T_6	Dimethoate 30 EC	2 ml/l
T_7	Control (water spray)	-

The pesticides were sprayed on the foliage of mulberry using hand sprayer in the field. Treatments were imposed once on 25th, 30th and 35th days after pruning (DAP). Polythene sheet barrier was used to avoid drifting of pesticides from one treated plot to another while spraying. The leaves from treated mulberry plants were harvested from 45 DAP onwards. The feeding of treated mulberry leaves was initiated from second feeding using third instar larvae.

C. Observations recorded

(i) Rearing parameters

Larval mortality (%). Larval mortality was recorded for all the treatments and the percent larval mortality was calculated.

Per cent larval mortality (%) =

$$\frac{\text{Number of dead larvae per treatment}}{\text{Total number of larvae per treatment}} \times 100$$

Per cent larval progression (%). The number of silkworms survived in different treatments upto spinning stage were recorded and the percent larval progression was calculated by using the formula,

Per cent larval progression =

$$\frac{\text{Total number of larvae alive per treatment}}{\text{Total number of silkworms per treatment}} \times 100$$

Instar duration (days /instar). The time taken by larvae to complete each instar was recorded by taking the total duration in days between two consecutive moults.

Maximum larval weight. The weight of ten randomly picked silkworms was recorded at third, fourth and fifth instars prior to moulting, for each replication and the average of the same was computed as maximum larval weight in each instar.

Effective Rate of Rearing (ERR) (%). The total number of cocoons harvested from each replication at the end of rearing were recorded and the ERR was calculated by using formula,

$$\text{ERR (\%)} = \frac{\text{Number of cocoons harvested}}{\text{Number of larvae brushed}} \times 100$$

D. Statistical analysis

The data was analyzed statistically for the test of significance using Fisher method of analysis of variance as outlined by Sundaraj *et al*. (1972). The population count and percentage values were subjected to square root and arc sine transformation, respectively. The interpretation of data was done using critical difference (CD) values. The Duncan's Multiple Range Test (DMRT) was conducted to compare the mean values among treatments was compared using CD value.

RESULTS AND DISCUSSION

A. Effect of chemical insecticides on larval mortality (%)

At 15 DAS, T2 (acetamiprid 20 SP @ 0.2 g / l) recorded highest mortality of 21.33 per cent followed by T4 (acephate 75 SP @ 1 g / l) (10.67 %), T7 (control) recorded lowest mortality of 0.67 per cent which is on par with T5 (imidacloprid 17.8 SL @ 0.3 ml / l) (1.33 %). However, in all other treatments larval mortality was not observed. Hence, 15 DAS would be the safe period for buprofezin 25 SC @ 2 ml / l, fipronil 5 SC @ 2ml / l and dimethoate 30 EC @2 ml / l.

At 20 DAS, T2 (acetamiprid 20 SP @ 0.2 g/l) recorded larval mortality of 10.00 per cent followed by T7 (control) (1.33%), while the other treatments did not exhibit anymortality. Therefore, the safe period for imidacloprid 17.8 SL @ 0.3 ml/l and acephate 75 SP @ 1 g/l would be 20 DAS (Table 1, Fig. 1). The highest mortality in acetamiprid might be due to presence of its residue evenup to 20 days after spray. Acetamiprid is a new molecule which shows systemic and trans-laminar action and it is a broad-spectrum insecticide having effect on both sucking pests as well as biting and chewing type of insects, this could be the reason for larval mortality even up to 20 DAS. Safety period is different for different insecticides, there will be no deleterious effect on silkworm growth and development on feeding insecticide treated leaves to silkworms after their respective waiting periods.

Under laboratory condition, Avramova *et al*. (2012) recorded the extent of toxicity of the neonicotinoid insecticides namely confidor and actara on silkworm hybrid Super-1 x Hesa-2. The larvae fed on insecticide treated leaves exhibited symptoms such as feeding rejection, shortening of the body, swelling, down shaking of the head, and vomiting of dark brown liquid after 15-20 minutes of feeding confidor treated leaves compared to 60-120 minutes with actarainsecticide

Table 1: Effect of chemical insecticides on larval mortality (%).

Treatments		Larval mortality (%)		
		10 DAS	15 DAS	20 DAS
T_1	Buprofezin 25 SC @ 2 ml / l	4.66	0.0	0.0
T_2	Acetamiprid 20 SP @ 0.2 g / l	24.67	21.33	10.00
T_3	Fipronil 5 SC @ 2 ml / l	6.66	0.0	0.0
T_4	Acephate 75 SP @ 1 g / l	15.33	10.67	0.0
T_5	Imidacloprid 17.8 SL @ 0.3 ml / l	5.33	1.33	0.0
T_6	Dimethoate 30 EC @ 2 ml / l	7.33	0.0	0.0
T_7	Control (water spray)	2.0	0.67	1.33
F-test		*	*	*
S.Em ±		2.553	0.959	0.471
CD @ 5 %		7.867	2.957	1.453

*Significant at 5%, DAS- Days after spraying

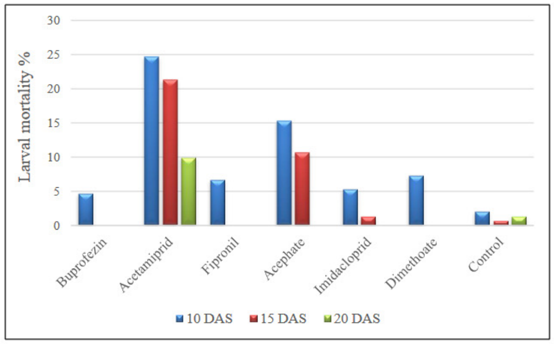
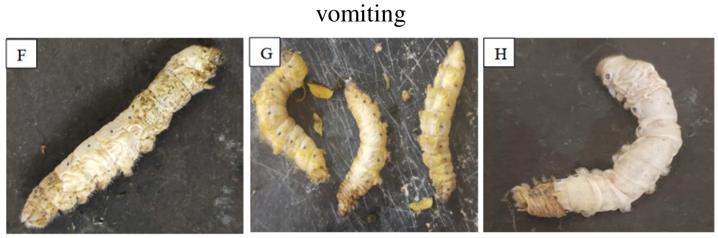


Fig. 1. Effect of chemical insecticides on larval mortality (%).

A. Rectal protrusion symptom exhibited by 4th & 5th instar larvae.

B. Vomiting gut juice with shrinkage of larval body.

C. Inactiveness, softening of the body, D. Death of the larvae without moulting, E. Twisting the body in agony and vomiting



F. Stiffening of the larval body, legs lose its clasping power, G. Death of ripened larvae without spinning, H. Incomplete moulting

Plate 1: Residual toxicity symptoms exhibited by acetamiprid 20 SP @ 0.2 g / l treated larvae.

B. Effect of chemical insecticides on larval progression (%) up to spinning stage

Larval progression (%) exhibited significant difference among treatments and among different days after spraying. At 15 DAS, 100 % larval progression was observed in T1 (buprofezin 25 SC @ 2 ml /l), T3 (fipronil 5 SC @ 2 ml / l) and T6 (dimethoate 30 EC @ 2 ml / l) and minimum was observed in T2 (acetamiprid 20 SP @ 0.2 g / l) (78.67 %) which was on par with T4 (acephate 75 SP @ 1 g / l) (89.33 %).

At 20 DAS, 100 % larval progression was observed in T1 (buprofezin 25 SC @ 2 ml / l), T3 (fipronil 5 SC @ 2 ml/l), T4 (acephate 75 SP @ 1 g / l), T5 (imidacloprid

17.8 SL @ 0.3 ml /l) and T6 (dimethoate 30 EC @ 2 ml / l) and minimum was observed in T2 (acetamiprid 20 SP @ 0.2 g / l) (90 %) and control recorded 98.67 % larval progression (Table 2, Fig. 2).

Similarly, fenpropathrin recorded decreased larval progression in both 15 and 20 DAS reflecting that pyrethroid group is not suitable for use in sericulture and can cause considerable damage to the growth and development in silkworms as it attacks the insect nerve membranes disrupting the nerve conduction (Fritz *et al.,* 2013).

C. Effect of chemical insecticides on larval weight (g/10 larvae)

Larval weight of silkworms fed on mulberry leaves treated with chemical insecticides at different intervals after spraying was recorded and the difference in larval weight between treatments were observed in all instars (third, fourth, and fifth).

Table 2: Effect of chemical insecticides on larval progression (%) up to spinning stage.

Treatments		larval progression (%)		
		10 DAS	15 DAS	20 DAS
T_1	Buprofezin 25 SC @ 2 ml / l	95.34	100.00	100.00
T_2	Acetamiprid 20 SP @ 0.2 g / l	75.33	78.67	90.00
T_3	Fipronil 5 SC @ 2 ml / l	93.34	100.00	100.00
T_4	Acephate 75 SP @ 1 g / l	84.67	89.33	100.00
T_5	Imidacloprid 17.8 SL @ 0.3 ml / l	94.67	98.67	100.00
T_6	Dimethoate 30 EC @ 2 ml / l	92.67	100.00	100.00
T_7	Control (water spray)	98.00	99.33	98.67
F-test		*	*	*
S.Em. ±		2.568	0.959	0.471
CD @ 5 %		7.912	2.957	1.453

*Significant at 5%, DAS- Days after spraying

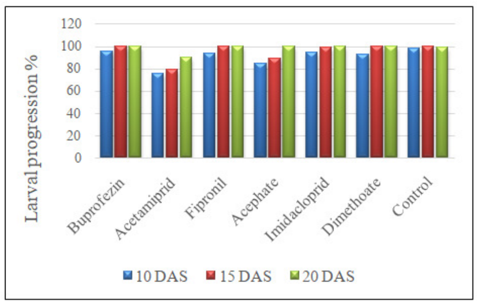


Fig. 2. Effect of chemical insecticides on larval progression (%) up to spinning stage.

Table 3: Effect of chemical insecticides on larval weight (g / 10 larvae.

Treatments	Larval weight (g / 10 larvae)								
	10 DAS			15 DAS			20 DAS		
	3^{rd} instar	4^{th} instar	5^{th} instar	3^{rd} instar	4^{th} instar	5^{th} instar	3^{rd} instar	4^{th} instar	5^{th} instar
T_1	0.86	12.22	25.87	0.91	12.69	26.30	0.91	12.70	26.63
T_2	0.78	9.02	21.42	0.81	9.96	22.33	0.57	10.08	23.40
T_3	0.85	12.08	26.10	0.88	12.11	26.17	0.88	12.26	26.77
T_4	0.84	10.19	24.10	0.85	11.11	24.87	0.81	11.65	25.57
T_5	0.85	12.13	26.00	0.89	12.61	26.64	0.92	12.90	27.30
T_6	0.88	12.55	29.08	0.96	12.90	29.80	1.10	13.17	29.33
T_7	0.92	13.04	30.15	1.04	13.23	30.17	1.24	13.41	30.43
F-test	*	*	*	*	*	*	*	*	*
S.Em. ±	0.012	0.303	0.348	0.032	0.416	0.327	0.027	0.130	0.336
CD @ 5 %	0.037	0.933	1.073	0.099	1.283	1.006	1.084	1.402	1.035

*Significant at 5%, DAS-Days after spraying

T1-Buprofezin 25 SC @ 2 ml / l T4- Acephate 75 SP @ 1 g/ l

T2- Acetamiprid 20 SP @ 0.2 g / lT5- Imidacloprid 17.8 SL @ 0.3 ml / l

T3- Fipronil 5 SC @ 2 ml / l T6- Dimethoate 30 EC @ 2 ml / l

T7- Control (water spray)

At 20 DAS, in 3rd instar the highest larval weight was recorded in T7 (control) (1.24 g / 10 larvae) followed by T6 (dimethoate 30 EC @ 2 ml / l) (1.10 g / 10 larvae), T5 (imidacloprid 17.8 SL @ 0.3 ml / l) (0.92 g / 10 larvae) and lowest weight was recorded in T2 (acetamiprid 20 SP @ 0.2 g / l) (0.57 g / 10 larvae) which was on par with T4 (acephate 75 SP @ 1 g / l) (0.81 g / 10 larvae). In 4th instar, the highest larval weight was recorded in T7 (control) which was 13.41 g / 10 larvae followed by T6 (dimethoate 30 EC @ 2 ml /

l) (13.17 g / 10 larvae), T5 (imidacloprid 17.8 SL @ 0.3 ml / l) (12.90 g / 10 larvae) and lowest weight was recorded in T2 (acetamiprid 20 SP @ 0.2 g / l) (10.08 g / 10 larvae) which was on par with T4 (acephate 75 SP @ 1 g / l) (11.65 g / 10 larvae). In 5^{th} instar, the highest larval weight was recorded in T7 (control) which was 30.43 g / 10 larvae followed by T6 (dimethoate 30 EC @ 2 ml / l) (29.33 g / 10 larvae), T5 (imidacloprid 17.8 SL @ 0.3 ml / l) (27.30 g / 10 larvae) and lowest was recorded in T2 (acetamiprid 20 SP @ 0.2 g / l) (23.40 g / 10 larvae) which was on par with T4 (acephate 75 SP @ 1 g / l) (25.57 g / 10 larvae) (Table 3). The variations could be due to the effect of feeding mulberry leaves treated with chemical insecticides. The residue of insecticides in mulberry leaves might have caused difference in larval weight (Plate 1).

Similarly, Sharma (1990) studied the relative contact toxicity of insecticides like BHC, nuvan, rogor, and dimecron, against grasshoppers, *Neorthacris acuticeps nilgiriensis*, as well as the persistence of these chemicals' toxicity to silkworm larvae. The toxicity of nuvan lasts only for one day, making it safer for silkworm rearing compared to other three pesticides applied. In several insect species, effect of pesticides ranged from delayed moulting with the development of malformation of insect bodyparts to the complete inhibition of growth at higher concentrations (Schmutterer, 1990). The first, second and third instar larvae of the silkworm *Bombyx mori* were severely affected by neem-based pesticides, which resulted in complete larval mortality and death of majority of the larvae without moulting (Jyothi *et al*., 1999)

D. Effect of chemical insecticides on larval duration (days)

Longer larval duration was observed at 15 DAS during 3^{rd}, 4^{th} and 5^{th}instar in T2 (acetamiprid 20 SP @ 0.2 g / l) which was 3.67, 4.96 and 8.17 days respectively followed by T4 (acephate 75 SP @ 1 g / l) (3.37, 4.67 and 7.57 days respectively) and shortest larval duration was recorded in T7 (control) (3.02, 4.06 and 7.17 days respectively) which was on par with T6 (dimethoate 30 EC @ 2 ml / l) (3.03, 4.04 and 7.32 days respectively).

At 20 DAS, during 3^{rd}, 4^{th} and 5^{th} instar, longer larval duration was observed in T2 (acetamiprid 20 SP @ 0.2 g / l) which was 3.73, 4.91 and 8.00 days respectively followed by T4(acephate 75 SP @ 1 g / l) (3.24, 4.67 and 7.63 days respectively) and shortest larval duration was recorded in T7 (control) (3.01, 4.00 and 7.13 days respectively) which was on par with T6 (dimethoate 30 EC @ 2 ml / l) (3.08, 4.13 and 7.20 days respectively (Table 4). The variations in the larval duration could be due to the residual effect of different chemicals.

In comparison to present study, Roxelle *et al*. (2013) studied the effect of chlorantraniliprole on silkworm larvae (Brazilian silkworm hybrids) showed that the highest percentage of mortality (100%) in larvae at 0.2 ppm, but less mortality was recorded (0.00 to 1.66%) at 0.025ppm. Even though lesser concentrations did not result in mortality, toxicity signs such as feeding cessation, regurgitation, delayed growth and incomplete ecdysis were seen. Earlier research has shown how pesticides affect *Bombyx mori* L. deal with toxicity, slowed or delayed growth and development, mortality, fecundity and food intake (Kuribayashi, 1988).

Table 4: Effect of chemical insecticides on larval duration (days).

Treatments	Larval duration (days)								
	10 DAS			15 DAS			20 DAS		
	3^{rd} instar	4^{th} instar	5^{th} instar	3^{rd} instar	4^{th} instar	5^{th} instar	3^{rd} instar	4^{th} instar	5^{th} instar
T_1	3.20	4.27	7.43	3.27	4.37	7.31	3.30	4.34	7.24
T_2	3.77	5.03	8.29	3.67	4.96	8.17	3.73	4.91	8.00
T_3	3.05	4.30	7.47	3.10	4.27	7.23	3.14	4.20	7.30
T_4	3.41	4.53	7.87	3.37	4.67	7.57	3.24	4.67	7.63
T_5	3.17	4.30	7.70	3.20	4.20	7.47	3.40	4.23	7.47
T_6	3.03	4.04	7.32	3.03	4.10	7.27	3.08	4.13	7.20
T_7	3.01	4.02	7.21	3.02	4.06	7.17	3.01	4.00	7.13
F-test	*	*	*	*	*	*	*	*	*
S.Em. ±	0.045	0.082	0.055	0.022	0.065	0.045	0.023	0.056	0.034
CD @ 5 %	0.139	0.252	0.171	0.069	0.200	0.120	0.070	0.173	0.104

*Significant at 5%, DAS-Days after spraying

T1-Buprofezin 25 SC @ 2 ml / l — T4- Acephate 75 SP @ 1 g/ l

T2- Acetamiprid 20 SP @ 0.2 g / l — T5- Imidacloprid 17.8 SL @ 0.3 ml / l

T3- Fipronil 5 SC @ 2 ml / l — T6- Dimethoate 30 EC @ 2 ml / l

T7- Control (water spray)

E. Effect of chemical insecticides on effective rate of rearing (ERR) (%)

Silkworm fed on mulberry leaves treated with chemical insecticides exhibited significant difference in ERR per cent among treatments at different days after spraying

At 15 DAS, maximum ERR percent was observed in T7 (control) (96.00 %) followed by T6 (dimethoate 30 EC @ 2 ml / l) (95.67 %), T1 (buprofezin 25 SC @ 2 ml / l) (95.33 %), T3 (Fipronil 5 SC @ 2 ml / l) (94.67 %), T5 (imidacloprid 17.8 SL @ 0.3 ml / l) (93.89 %) and least ERR % was recorded in T2 (acetamiprid 20 SP @ 0.2 g / l) (57.12 %) and T4 (acephate 75 SP @ 1 g / l) (87.30 %).

At 20 DAS, maximum ERR percent was observed in T7 (control) (98.00 %) and T6 (dimethoate 30 EC @ 2 ml / l) (98.00 %) followed by T1 (buprofezin 25 SC @ 2 ml / l)(96.67 %) and T5 (imidacloprid 17.8 SL @ 0.3 ml / l) (96.67 %), T3 (Fipronil 5 SC @ 2 ml / l) (96.64 %) and least ERR % was recorded in T2 (acetamiprid 20

SP @ 0.2 g / l) (68.89 %) and T4 (acephate 75 SP @ 1 g / l) (95.33 %) (Table 5).

The differed ERR % among treatments could be due to the mortality of silkworms due to insecticidal residue in respective treatments.

Similarly, Yeshika *et al*. (2019) studied the effects novel insecticides with distinct mode of action when applied as foliar spray to mulberry leaves. The results showed that flonicamid 50 WG @ 0.3 g/l recorded 100per cent silkworm mortality. Maximum larval weight was recorded in dinotefuran 20 SG @ 0.25 g/l and dichlorvos 76 EC @ 2.63 ml/ l at 10, 20 30, 40 DAS. 100 per cent ERR %was recorded in pymetrozine 50 WG @ 0.3 g/l at 10 DAS, azadirachtin 1% @ 2 ml/l and dichlorvos 76 EC @ 2.63 ml/l at 30 DAS.

Table 5: Effect of chemical insecticides on effective rate of rearing (ERR) (%).

Treatments		Effective Rate of Rearing (ERR %)		
		10 DAS	15 DAS	20 DAS
T_1	Buprofezin 25 SC @ 2 ml / l	91.92	95.33	96.67
T_2	Acetamiprid 20 SP @ 0.2 g / l	56.93	57.12	68.89
T_3	Fipronil 5 SC @ 2 ml / l	89.17	94.67	96.64
T_4	Acephate 75 SP @ 1 g / l	68.68	87.30	95.33
T_5	Imidacloprid 17.8 SL @ 0.3 ml / l	90.15	93.89	96.67
T_6	Dimethoate 30 EC @ 2 ml / l	88.39	95.67	98.00
T_7	Control (water spray)	92.58	96.00	98.00
F-test		*	*	*
S.Em. ±		2.603	1.873	0.822
CD @ 5 %		8.020	5.770	2.532

CONCLUSIONS

Acetamiprid belongs to neonicotinoid group of chemicals which shows systemic and translaminar action and it is also belongs to broad-spectrum insecticide, which is mainly used to control sucking pests but it also shows its effect on biting and chewing type of insects hence its residue on mulberry leaves showed adverse effect on silkworm performance with toxicity symptoms like rectal protrusion, vomiting gut juice with shrinkage of larval body, twisting the body in agony and stiffening of the larval body *etc.* From this study it is suggested that fipronil and dimethoate had a reasonably good safety period of 15 days and resulted in better rearing parameters.

FUTURE SCOPE

Safety period of acetamiprid was found to be more than 20 DAS therefore the exact safety period needs to be established.

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