ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Bio-Efficacy and Dissipation of Newer Molecules Against White Fly in Okra.

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ABSTRACT

Experiments were conducted during *kharif,* 2013 to evaluate the efficacy of six insecticides *viz.,* bifenthrin 10 EC at 80 g a.i. ha^{-1}, fipronil 5 SC at 500 g a.i. ha^{-1}, flubendiamide 480 SC at 60 g a.i. ha^{-1}, quinalphos 25 EC at 350 g a.i. ha^{-1}, profenofos 50 EC at 400 g a.i. ha^{-1} and beta-cyfluthrin 25 SC at 18.75 g a.i. ha^{-1} against white fly *Bemisia tabaci* (Gennadius) on okra of which flubendiamide @ 60 g a.i./ha, profenofos @ 400 g a.i./ha and beta-cyfluthrin @ 18 g a.i./ha, applied twice as foliar sprays were found to be most effective in controlling the okra pests, hence the dissipation studies were conducted for these three insecticides. The dissipation pattern was studied by collecting okra fruits at 0, 1, 3, 5, 7, 10 and 15 days after last spray and analyzed at AINP on Pesticide Residues, Rajendranagar, Hyderabad The initial deposits of flubendiamide at 60 g a.i. ha^{-1} sprayed at 50 % flowering and repeated 15 days after first spray recorded 1.49, mg kg^{-1} which dissipated to below detectable level (BDL) on 10th day. The initial deposit of profenofos (400 g a.i./ha) in okra fruits was 1.52 mg/kg which degraded to 0.09 by 7th day after last spraying. The initial deposit of beta-cyfluthrin (18.75 g a.i./ha) in okra fruits was 0.11 mg/kg and by one day after treatment dissipated by 45.04 per cent.

Keywords: Okra, insecticides, whitefly , initial deposit, efficacy, dissipation and half-life.

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INTRODUCTION

Vegetables constitute an important item of our food supplying vitamins, carbohydrates and minerals needed for a balanced diet and of high value specially in under developed and developing countries like India, where malnutrition abounds (Masood Khan *et al.,* 2001). Okra, *Abelmoschus esculentus* (L.) Moench is one of the major economically important vegetable crops grown in India in an area of 0 .451 m ha with an annual production of 4.796 m mt and productivity of 10.62 ton per hectare (CMIE, 2011) accounting for 21 per cent of total exchange earnings from export of vegetables from India grown mostly in Assam, Uttar Pradesh, Bihar, Orissa, West Bengal, Maharashtra, Andhra Pradesh and Karnataka. Besides various other factors for lower productivity, heavy damage is inflicted by major insect pests *viz.,* leafhopper, *Amrasca biguttula biguttula* (Ishida), whitefly, *Bemisia tabaci* (Gennadius) and shoot and fruit borer, *Erias vittella* (Fabricius) . Among these, whitefly, *Bemisia tabaci* (Gennadius) is a serious pest , known to be the vector of vein clearing disease (Gupta et al. 2009). Indiscriminate use of pesticides leads to undesirable load of pesticide residues in marketable vegetables (Kumari *et al.*,2005) and cause severe ecological consequences like destruction of natural enemy fauna, effect on non-target organisms, and directly affect in the form of residues. Hence studies were conducted to evaluate the efficacy of different insecticides used commonly and also to establish the dissipation pattern of effective insecticides to fit in pest management strategy.

MATERIALS AND METHODS

Okra crop was raised in randomized block design with seven treatments replicated thrice using "Arka Anamika" variety at spacing of 45×15 cm and the insecticides Bifenthrin 10 EC at 80 g a.i. ha^{-1}, fipronil 5 SC at 500 g a.i. ha^{-1}, flubendiamide 480 SC at 60 g a.i. ha^{-1}, quinalphos 25 EC at 350 g a.i. ha^{-1}, profenofos 50 EC at 400 g a.i. ha^{-1} and beta-cyfluthrin 25 SC at 18.75 g a.i. ha^{-1} were sprayed on okra plants at 50 % of flowering and thereafter, repeated at 15 days interval using knapsack sprayer .For the efficacy studies the population of whitefly were recorded on five randomly selected plants per plot leaving the border rows. The population counts were recorded from top, middle and bottom leaf in each of the five selected plants in every plot and mean number of whiteflies per five plants were calculated. Then data was analyzed with arc sine values obtained from the conversion of percentage of infestation of fruits (Gomez and Gomez, 1984).The percentage reduction at one, three, five, seven and ten days after each spraying were pooled and transformed into arc sine values which were further subjected to statistical analysis. The overall effect of the treatments by combining these five observations were also assessed by analyzing the data thorough ANOVA.

Dissipation

Three most effective insecticidal treatments from the efficacy studies viz., flubendiamide at 60 g a.i. ha-1, profenofos at 400 g a.i. ha-1 and beta-cyfluthrin at 18 g a.i. ha-1 were utilized for residue analysis study. The okra fruits samples of 250 g were collected randomly from each plot at 0, 1, 3, 5, 7, 10 and 15th day after last spray in polythene bags and brought to the laboratory immediately for further sample processing and further procedures adopted for dissipation studies in the laboratory and analysed on HPLC and GC for residue determination.

Extraction and Clean-up for flubendamide

Representative sample (okra fruits) of 50 g was extracted with 50 ml acetonitrile twice by using mechanical shaker for 30 minutes. The extraction mixture was filtered and evaporated to near dryness using a vacuum rotary evaporator and the contents were re dissolved in 40 ml of acetonitrile. A glass column was packed with 5 g of alumina using hexane as solvent and drained the excess of solvent. The sample was transferred in to the column and eluted with 100 ml of 10:1 hexane: ethyl acetate solvent mixture. Discarded the first 10 ml fraction and collected the elute over anhydrous sodium sulphate, the process was repeated thrice. Approximately 300 ml of elute was collected. The elute was completely dried or drained and evaporated to near dryness. The residues were recovered in 5 ml volume of acetonitrile for High Pressure Liquid Chromatograph (HPLC) analysis.

Extraction and clean up for Profenofos

Representative sample (okra fruits) of 25 g was homogenized with 50 ml acetontrile and was filtered. Then filtrate was evaporated to near dryness using a vacuum rotary evaporator and the contents were re dissolved in 25 ml of hexane. Then recovered filtrate was partitioned after adding 100 ml of acetonirile and 125 ml of 5 per cent sodium chloride solution. The extract was cleaned up with florosil column eluting with dichloromethane. The elute was concentrated again and dissolved in n-hexane and later subjected to alumina column clean up. The final elution was done with hexane: acetone (9:1) and evaporated to dryness and analyzed on Gas Chromatograph (GC).

Extraction and Clean-up for Beta-cyfluthrin

Representative fruit sample of 25 g was homogenized with 50 ml acetone:hexane (1:9) and was filtered. The filtrate was partitioned after adding with saturated Nacl and Dichloromethane. The extract was cleaned up with florosil column eluting with hexane. The elute evaporated to dryness for Gas Chromatography analysis.

Estimation

The residues flubendamide were estimated by using HPLC –PDA and beta-cyfluthrin, profenofos with GC-ECD by comparing peak area of the standard with that of peak in the sample under identical conditions.

From the technical grade of flubendamide, betacyfluthrin and profenofos, one ppm standard solution was prepared by diluting with n-hexane and used for carrying out recovery and comparative studies of pesticide residues in the fruit samples collected at different intervals.

The recovery study of flubendiamide was carried out at the levels of 0.01, 0.10 in okra fruit Recoveries ranged from 86–88 respectively with standard deviation of 2.15–6.58%. The recovery study of Profenophos was carried out at the levels of 0.01, 0.10 in okra fruit, Recoveries were ranged from 86–88 % respectively with standard deviation of 1.12–4.57%. The recovery study of Beta cyfluthrin carried out at the levels of 0.01, 0.10 in okra fruit, and recovery ranged from 87–88 % respectively with standard deviation of 1.10–3.28%.

Flubendiamide sample analyses were done with Shimadzu HPLC equipped with PDA detector. A reverse-phase RP-18e, Chromolith 100 x 4.6 mm i.d (10 mm), 150 mm long was used as the column and maintained at room temperature. The mobile phase consisted of acetontirile/water (60/ 40, v/v), with a flow rate of 0.5 mL min-1 up to 15min by the mobile phase of acetontirile/water (80/ 20, v/v), then 15.01 min to 25 min with a flow rate of 0.5ml/min by the mobile phase of acetontirile/water (60/ 40, v/v). The retention time (RT) was 8.97 min for flubendiamide.

The Profenophos and Beta cyfluthrin sample were analyzed on (Shimadzu) GC-2010 equipped with fused silica capillary column Factor Four (30 mt×0.25 mm id) coated with 1% phenyl-methylpolysiloxane (0.25 µm film thickness) using ^{63}Ni electron-capture detector (ECD). General operating conditions were as follows: For Profenophos ,Column temperature program: initially 80°C for 5 min, increase at 2◦C/min to 150°C hold for 10min, then 280◦C increase 2◦C/min hold for 2min, Total programme is 93.00 min; injection volume: 1 µl nitrogen flow rate 0.93 ml/min and makeup 25 ml/min with split ratio 1:10; using carrier gas (N2) 99.5%; Injector port temperature 260°C; detector temperature 300°C. Retention time of Profenophos is 48.9min. For Beta cyfluthrin Column temperature program: initially 200°C for 2 min, increase at 3◦C/min to 240°C hold for 10min, Total programmes is 25.33 min; injection volume: 1 µl nitrogen flow rate 0.93 ml/min and makeup 25 ml/min with split ratio 1:10; using carrier gas (N2) 99.5%; Injector port temperature 260°C; detector temperature 300°C. Retention time of Beta cyfluthrin is is 14.9 min

RESULTS AND DISCUSSION

Bioefficacy results against white fly (Table-1) show that Beta-cyfluthrin at 18.75 g a.i./ha, quinalphos at 350 g a.i./ha and bifenthrin at 80 g a.i./ ha recorded high per cent reduction of white fly population of 54.21, 54.21 and 50.69 per cent, respectively compared to fipronil at 500 g a.i./ha (46.62%), profenofos at 400 g a.i./ha (40.73%) and flubendiamide at 60 g a.i./ha (33.05%) after second spray. The observations on over all efficacy revealed that all the insecticidal treatments were superior to control. Beta-cyfluthrin at 18.75 g a.i./ha and quinalphos at 350 g a.i./ha were most effective and significantly superior to all other treatments recording 57.00% and 54.42% of white fly population reduction, respectively. The other promising treatments were bifenthrin at 80 g a.i./ha (51.69%) and profenofos at 400 g a.i./ha (51.34%) which were superior over other treatments and were on par with beta-cyfluthrin and quinalphos. Fipronil at 500 g a.i./ha and flubendiamide at 60 g a.i./ha were least effective among all insecticides tested with population reduction of 47.28 and 46.42 per cent respectively(figure 1). The observations recorded after second spray with regard to the reduction of whitefly population indicated that all the treatments were significantly superior to control. The efficacy in descending order was beta-cyfluthrin at 18.75 g a.i./ha, quinalphos at 350 g a.i./ha, bifenthrin at 80 g a.i./ha, fipronil at 500 g a.i./ha and profenofos at 400 g a.i/ha with reduction of 54.21, 54.21, 50.69, 46.62 and 46.14 per cent, respectively. All the insecticides except flubendiamide at 60 g a.i./ha were superior over in controlling white fly population. Flubendiamide at 60 g a.i./ha exhibited least white fly population reduction of 37.35 per cent.(figure 2). But very efficient in the reduction of lepidopterous insects (Hirooka etal 2007, Tohnishi etal 2005, Nauen etal 2007) and fruit and shoot borer (Jagginavar etal 2009). The overall mean efficacy showed that among all the treatments, beta-cyfluthrin at 18.75 g a.i./ha is the most effective in controlling white fly population.It is also found that quinalphos at 350 g a.i./ha was promising against white fly population followed by bifenthrin at 80 g a.i./ha. The findings of the present study proved that profenofos at 400 g a.i. ha-1 is an effective insecticide in controlling the whitefly.

Table 1: Efficacy of insecticides against white fly. *Bemisia tabaci* Gennadius. After 2 sprays

Treatment	Dosage (g a.i./ha)	Mean % of reduction over untreated check	
		Over all after 1st spray	**Over all after 2nd spray**
T_1 Bifenthrin	80	51.69^{ab} (46.0)	50.69^{a} (45.4)
T_2 Fipronil	500	47.28^{b} (43.4)	46.62^{a} (43.0)
T_3 Flubendiamide	60	46.42^{b} (42.9)	37.35^{b} (37.6)
T_4 Quinalphos	350	54.42^{a} (47.5)	54.21^{a} (47.4)
T_5 Profenofos	400	51.34^{ab} (45.8)	46.14^{a} (42.8)
T_6 Beta-cyfluthrin	18.75	57.00^{a} (49.0)	54.21^{a} (47.4)
T_7 Control	--	0.00 (0.00)	0.00 (0.00)
S.Em	--	1.14	1.62
C.D at 5%	--	3.63	5.18

DAS - Days After Spraying.
Figures in the parentheses are angular transformed values.

Table 2: Dissipation of Flubendiamide , Profenofos and beta-cyfluthrin in okra

Days	Flubendamide residues	Dissipation %	Profenfos residues	Dissipation %	beta-cyfluthrin residues	Dissipation %
0	1.49	0	1.52	0	0.11	0
1	0.84	43.30	1.14	24.92	0.06	45.40
3	0.47	68.08	0.75	50.44	0.02	80.95
5	0.10	93.14	0.62	58.96	BDL	100
7	0.01	98.88	0.09	93.88	BDL	100
10	BDL	100	BDL	100	BDL	100
15	BDL	100	BDL	100	BDL	100

*BDL- Below the Detectable level

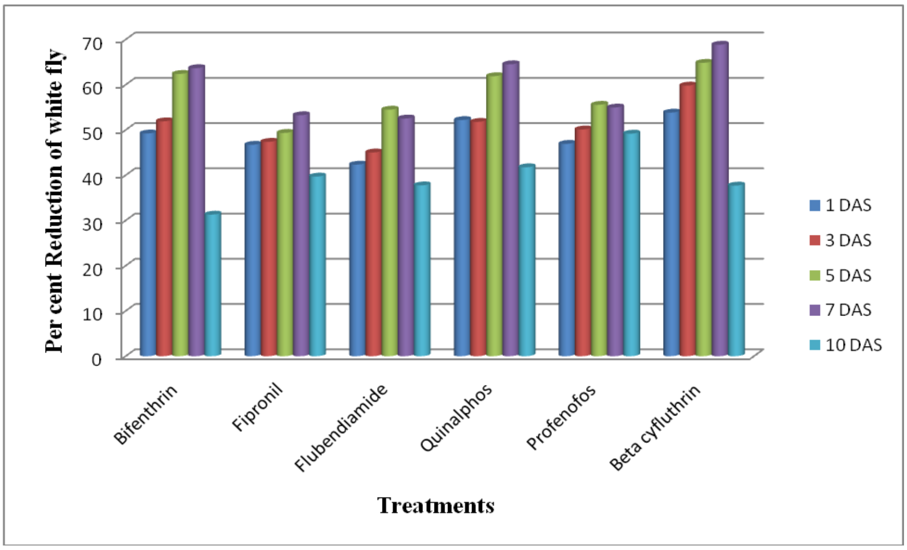

Figure 1: Efficacy of insecticides against whitefly, *Bemisia tabaci* (Gennadius) after first spray

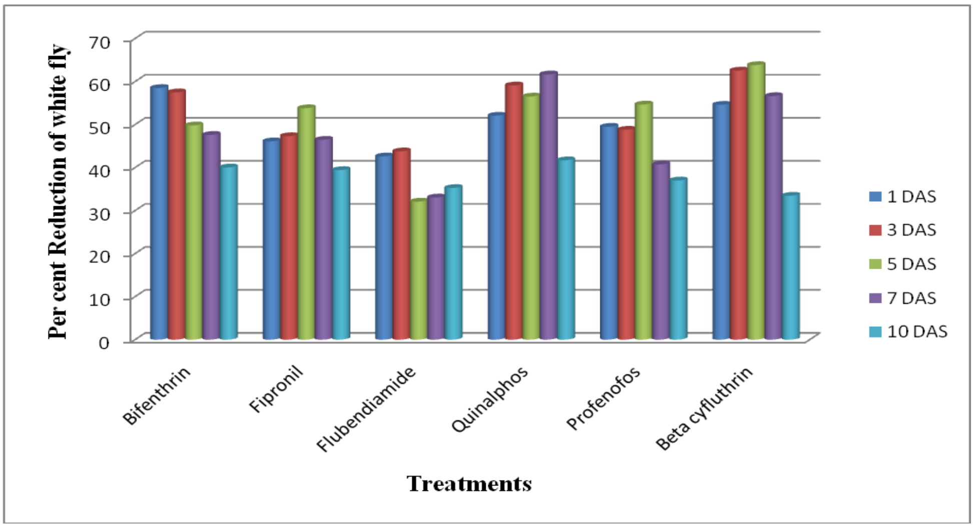

Figure 2: Efficacy of insecticides against whitefly, *Bemisia tabaci* (Gennadius) after second spray

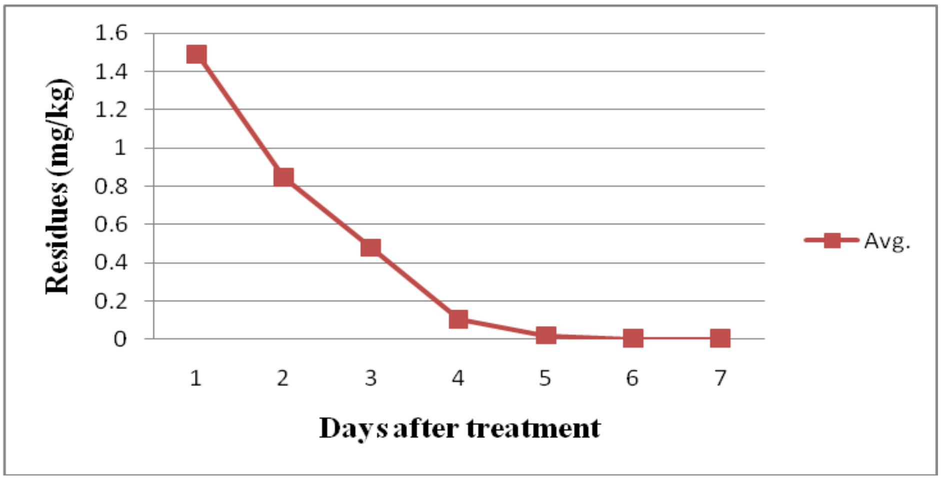

Figure 3: Dissipation of flubendiamide (60 g a.i./ha) in okra

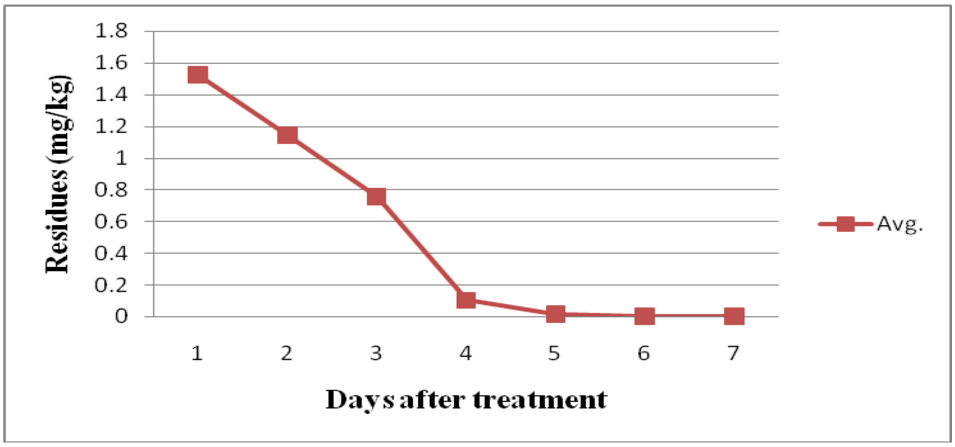

Figure 4: Dissipation of profenofos (400 g a.i./ha) in okra

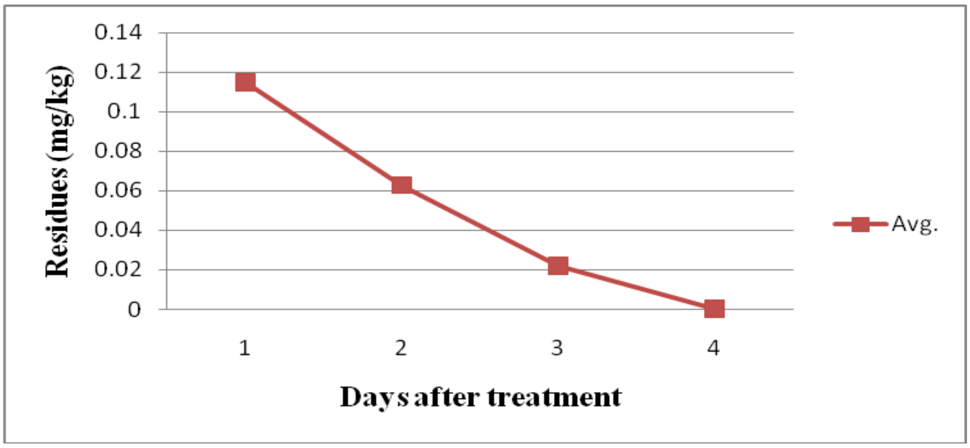

Figure 5: Dissipation of beta-cyfluthrin (18.75 g a.i./ha) in okra

Dissipation

The initial deposit and subsequent residues of flubendiamide (60 g a.i. ha^{-1}) in okra fruits at an interval of 0, 1, 3, 5, 7, 10 and 15 days after last spray are presented in table -2 and in figure 3 and chromatograms presented in figure 4. An initial deposit of 1.49 mg kg^{-1} gradually dissipated to 0.84, 0.47, 0.10 and 0.01 mg kg^{-1} at 1, 3, 5 and 7 days, respectively. The per cent dissipation was 43.30, 68.08, 93.14 and 98.88, respectively. The residues fell below maximum residue limit (MRL) of 0.2 mg kg^{-1} in 4.19 days after the treatment. The half-life (RL_{50}) of flubendiamide was worked to be 1.83 days. The present findings are in agreement with observations of Sahoo *et al.* (2009), who reported an initial deposit of 1.06 and 2.00 mg kg^{-1} following application of flubendiamide 480 SC at 60 and 120 g a.i. ha^{-1} and which were dissipated to below detectable level of 0.01 mg kg^{-1} in 7 and 10 days at single and double dosages, respectively. Das *et al*. (2011) reported that in okra the initial deposits of 0.28 and 0.53 µg g^{-1} reached below determination level (BDL) of 0.01 µg g^{-1} on the 7th and 10th day with half-life 4.7-5.1 days when flubendiamide 39.35 SC sprayed at 24 and 48 g a.i. ha^{-1}, respectively. The variation of initial deposits in the present findings with Das *et al*. 2011 may be due to variation in the dosages applied on the crop. Chawla *et al.* (2011) reported that residues of flubendiamide reached below determination level on 5^{th} and 10^{th} day after application at 90 and 180 g a.i. ha^{-1}, respectively on brinjal. (Sahoo,S.K 2009) explained in Dissipation kinetics of flubendiamide on chillis. The variation in the rate of dissipation in okra, brinjal and chillies may be due to changes in the crop matrix.

The initial deposit and subsequent residues of Profenofos (400 g a.i./ha) in okra fruits at intervals of 0, 1, 3, 5, 7, 10, 15 days after last spraying Table-2 have shown an initial deposit of 1.52 mg/kg which degraded to 0.09 by 7^{th} day after last spraying on okra.. The dissipation pattern showed constant decrease of residues from first day to 7^{th} day .The residues dissipated to 24.92, 50.44, 58.96 and 93.88 per cent by 1, 3, 5 and 7^{th} day respectively (Fig-4). and reached below tolerance limit of 0.5mg/kg in 5.1days with half-life

2.16.days.The initial deposit and subsequent residues of beta-cyfluthrin (18.75 g a.i./ha) in okra fruits at intervals of 0, 1, 3, 5, 7, 10, 15 days after last spraying (Table-2) have shown the initial deposit as 0.11 mg/kg and by one day after treatment dissipated by 45.04 per cent. Further very rapid dissipation was evident by third day dissipated to below tolerance limit of 0.02 mg kg-1 in 2.62 days.(Fig5)

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