Dissipation and degradation kinetics of commonly used pesticides and their metabolites in/on okra, *Abelmoschus esculentus* (L.) Moench

C. Meenambigai[1,*] and K. Bhuvaneswari[2]

[1]Department of Agricultural Entomology, Bidhan Chandra Krishi Viswavidyalaya, Nadia 741 252, India
[2]Department of Agricultural Entomology, Tamil Nadu Agricultural University, Coimbatore 641 003, India

Based on the dissipation pattern and degradation kinetics study of pesticides in okra, the average initial deposit of dimethoate and acephate was comparatively higher than emamectin benzoate and flubendiamide. Acephate residues persisted much longer, while, emamectin benzoate persisted for a shorter time. Acephate metabolized to methamidophos on 1 day after treatment. Des-ido flubendiamide residues were not detected. Okra being harvested on alternate days, following a pre-harvest interval of 24 days after acephate application, is not possible. Thus, usage of acephate at the flowering stage in okra poses the risk of residue detection in the harvested produce. Following a pre-harvest interval of 3–11 days after spraying dimethoate, emamectin benzoate and flubendiamide are mandatory for the safe consumption of okra.

Keywords: Acephate, dissipation, dimethoate, emamectin benzoate, half-life, okra.

ECONOMICALLY significant vegetable crops like okra (*Abelmoschus esculentus* (L.) Moench) are widely grown in India. Insect pests are an important constraint in okra production. The yield loss caused by pests such as leaf hopper, whitefly, aphids, two-spotted spider mite, and fruit and shoot borer is 32.06% to 56% (ref. 1), 94.02% (ref. 2), 54.04% (ref. 3), 17.46–48% (ref. 4) and 36–90% (ref. 5) respectively. To overcome such economic loss, farmers began the indiscriminate usage of pesticides. As a result, the detection of pesticide residues in harvested produce above the maximum residual limit (MRL) is emerging as one of the major bottlenecks in international trade. Pesticide residue detection was recorded in around 18.70% vegetables, of which 1.90% samples exceeded MRL fixed by the Food Safety Standards Authority of India (FSSAI) and 13.30% samples were found to have residues of non-approved pesticides[6]. The pesticide residue detection was maximum in retail outlets as well as in market vegetables such as brinjal, tomato and okra[6]. Based on the extensive survey conducted among okra farmers in Tamil Nadu, India, for insect pest control, the use of a wide range of pesticides from organophosphorus (OP) compounds to newer generation pesticides such as acephate 75 SP, dimethoate 30 EC, emamectin benzoate 5 SG and flubendiamide 480 SC was reported[7]. Dimethoate and acephate belong to OP, the most widely used group of pesticides and both fall under the moderately hazardous category of toxicity class II (refs 8 and 9). According to the Central Insecticide Board and Registration Committee (CIB and RC), Government of India (GoI), the usage of acephate on okra is not approved[10]. Around 5–10% of acephate applied in plants gets metabolized to methamidophos. This metabolite belongs to the highly hazardous category of toxicity class Ib[9,11–13]. Emamectin benzoate is synthetically derived from abamectin. It belongs to the moderately hazardous toxicity category of toxicity class II (ref. 9). One of the benzene dicarboxamide group of pesticides is flubendiamide, which is slightly hazardous in nature[9,14]. In crop and soil surfaces as well as in aqueous solution, during photodegradation of flubendiamide, iodine moiety is lost from the parent molecule to form a significant transformation product called des-iodo flubendiamide[15]. Both emamectin benzoate and flubendiamide are new-generation pesticides approved for use on okra against fruit and shoot borer in India. The present study was undertaken under tropical climatic conditions of Tamil Nadu to determine the dissipation pattern and degradation kinetics as well as fix the safe harvest interval of non-approved, widely used pesticides like acephate and methamidophos as well as approved pesticides like dimethoate, emamectin benzoate, flubendiamide and des-ido flubendiamide in okra.

Materials and methods

Chemicals and reagents

Sigma Aldrich (Bangalore, India) supplied the reference standards of dimethoate (99.5% purity), acephate (97.8% purity) and methamidophos (98.5% purity), emamectin benzoate (99.3% purity), flubendiamide (98.1% purity) and des-iodo flubendiamide (99.2% purity). Formulations such as dimethoate 30 EC (Rogar®), acephate 75 SP (Starthene®), emamectin benzoate 5 SG (Proclaim®) and flubendiamide 480 SC (Fame®) were procured from a local market. Merck

*For correspondence. (e-mail: meenaento@gmail.com)

(Mumbai, India) provided solvents such as HPLC-grade acetonitrile and hexane, MS-grade acetonitrile and reagents (AR grade) like anhydrous sodium sulphate, sodium chloride and anhydrous magnesium sulphate. Agilent Technologies (Chennai, India) supplied graphitized carbon black (GCB) and primary secondary amine (PSA).

Standard solutions

In a volumetric flask (25 ml), 24.60, 26.50, 33.50, 29.50, 26.75 and 25 mg reference standards of dimethoate, acephate, methamidophos, emamectin benzoate, flubendiamide and des-ido flubendiamide respectively was added separately. For 25 ml primary stock (1000 mg l^{-1}) of dimethoate, acephate and methamidophos, the volume was made up to the mark with hexane (HPLC-grade). While for emamectin benzoate, flubendiamide and des-ido flubendiamide, acetonitrile (HPLC-grade) was used. Working standard solutions of dimethoate, acephate and methamidophos (0.01–1 mg l^{-1}), emamectin benzoate, flubendiamide and des-ido flubendiamide (0.01–0.1 mg l^{-1}) were made by subsequent dilution of secondary stock (10 mg l^{-1}) with their respective solvents.

Field experiment

A field trial was conducted at Narasipuram village (10.988°N, 76.776°E) of Coimbatore, Tamil Nadu. Okra seeds (CO-4 hybrid) were sown at 45 × 30 cm spacing and grown according to Tamil Nadu Agricultural University (TNAU) – proposed cultivation practice (except for pest management). A randomized block design was used and treatments were replicated thrice along with untreated control in a 100 sq. m plot per replication, with 2 m alley spacing between the replications. The okra plots were sprayed with dimethoate 30 EC at 300 g a.i. ha^{-1}, acephate 75 SP at 292 g a.i. ha^{-1}, emamectin benzoate 5 SG at 8.5 g a.i. ha^{-1} and flubendiamide 480 SC at 48 g a.i. ha^{-1}. Two sprayings were done using a 500 l ha^{-1} capacity hand-operated knapsack sprayer, one at 50% flowering stage and the other 10 days after the first spraying. The experiment was conducted after obtaining permission from the respective farmers who owned the experimental sites.

Sampling, extraction and clean-up

From the experimental plots, okra fruits (1 kg) were picked at 0 (1 h after the second spraying), 1, 3, 5, 7, 10 and 15 days. In the laboratory, using a silent crusher M homogenizer, the samples were blended for 4 min at 14,000 rpm. QuEChERS procedure with the following modification was used for residue analysis in okra[16]. In a centrifuge tube (50 ml), 10 g sample and 20 ml acetonitrile were added and swirled for 1 min in a rotospin. Sodium chloride and anhydrous magnesium sulphate of 1 and 4 g respectively, were incorporated into the mixture, swirled and subsequently subjected to centrifugation for 10 min at 6000 rpm. Anhydrous sodium sulphate was taken in a test tube and the supernatant (9 ml) was poured into it. A centrifuge tube (15 ml) prefilled with 600, 100 and 10 mg of anhydrous magnesium sulphate, PSA and GCB respectively, was taken. To this, supernatant (6 ml) was added, swirled for 1 min and subjected to centrifugation for 10 min at 3000 rpm. The resultant supernatant (4 ml) was put in another test tube and eventually dried by means of nitrogen flow at 40°C in a Turbovap. For dimethoate, acephate and methamidophos, 1 ml hexane (HPLC-grade) was added into a dried test tube, mixed well, transferred to an autosampler vial and analysed using gas chromatography (GC). Instead of hexane, acetonitrile (HPLC-grade) was used for flubendiamide, emamectin benzoate and des-ido flubendiamide. Their residues were quantified using ultra high-performance liquid chromatography (UHPLC).

Instrumental parameters

GC-FPD: Dimethoate, acephate and methamidophos residues were estimated using GC (Shimadzu 2010). The instrument has an injector (Shimadzu AOC 20i), flame photometric detector (FPD) and autosampler (Shimadzu AOC 20S). DB-1 capillary column of 30 m length, 0.25 mm inner diameter and 0.25 µm particle size was used. The oven temperature was programmed initially at 130°C (0 min), increased to 250°C (1 min) and then again increased to 280°C (0 min). The injection volume was 2 µl operated in splitless mode. The detector as well as the injector was maintained at 280°C during operation. Carrier gas nitrogen flowed at 0.93 ml min^{-1} speed. In FPD, hydrogen and zero air had a flow rate of 90 and 120 ml min^{-1} respectively. The total run time of the GC programme was 15 min.

UHPLC: The residues of emamectin benzoate, flubendiamide and des-ido flubendiamide were estimated using UHPLC (Shimadzu, i series 2020). The instrument has an autosampler, C18 column (Agilent) and photodiode array detector. The column operated at 40°C was of 250 mm length, 4.6 mm inner diameter, 5 µm particle size and set at reverse phase. A mixture of 5 mM ammonium acetate and MS-grade acetonitrile (50 : 50) was used as the mobile phase at a low-pressure gradient. The flow rate, injection volume and total run time were 1 ml min^{-1}, 20 µl and 15 min respectively. The instrumental parameters were controlled using LC Solutions software.

Method validation

Based on SANTE guidelines, validation of the analytical method used for residue estimation was done[17]. Using five linear levels of the working standard solution, linearity was studied as triplicates in GC-FPD (0.01 to 1 mg l^{-1}) and

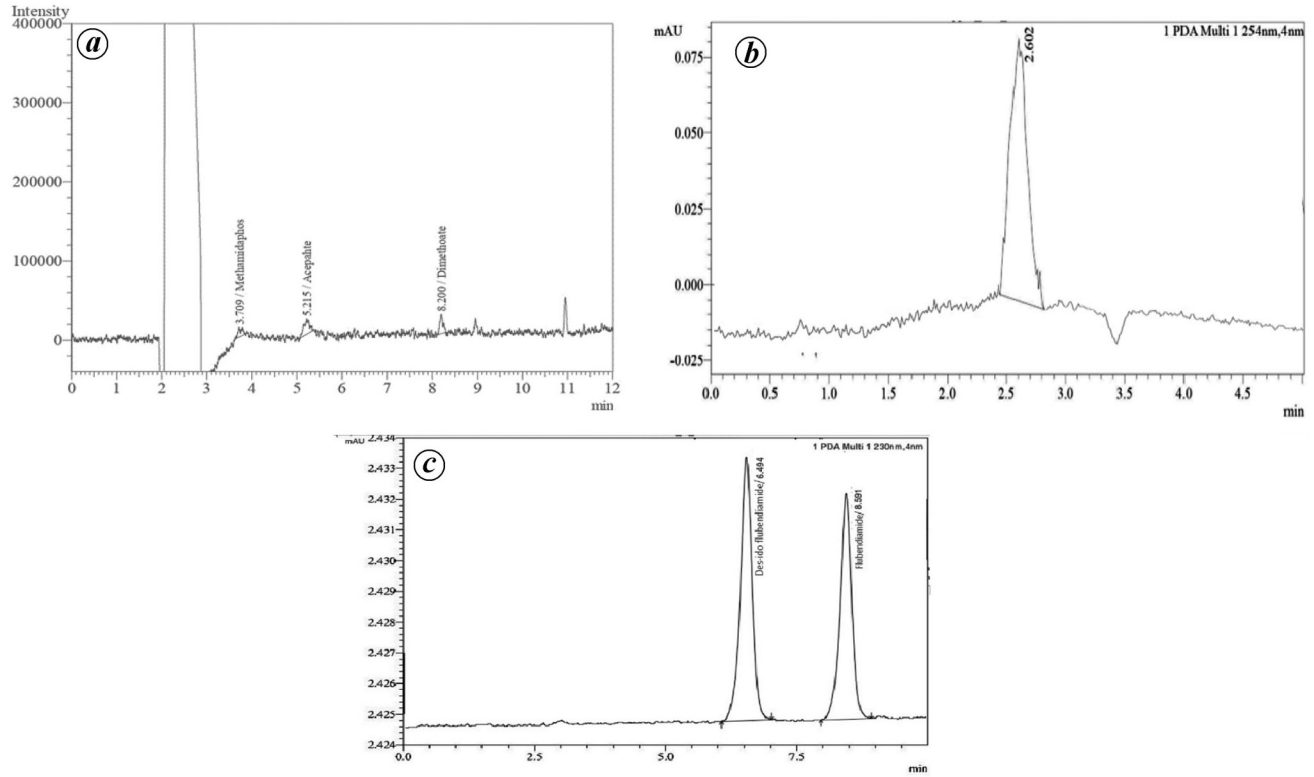

Figure 1. Chromatogram of okra sample spiked at limit of quantification (LOQ) level (0.01 mg kg^{-1}): (***a***) GC – dimethoate, acephate and methamidophos (organophosphorus (OP) mix), (***b***) UHPLC – emamectin benzoate and (***c***) UHPLC – flubendiamide and des-ido flubendiamide.

UHPLC (0.01 to 0.1 mg l^{-1}). Matrix effect (ME) based on a comparison of analyte response prepared in solvent to that prepared in the blank matrix of okra fruit (matrix-matched standard) was assessed[18]. On the basis of signal : noise of 3 : 10 with reference to the background noise of the blank sample, limit of detection (LOD) and limit of quantification (LOQ) were determined. For recovery studies, untreated homogenized samples (10 g) were treated with 0.01, 0.05 and 0.1 mg kg^{-1} of working standard solution and three replications were maintained along with control. Treated samples were kept undisturbed for 1 h and residues were estimated using the above-mentioned modified QuChERS method. Precision of the analytical method was determined through repeatability for each treated concentration (0.01, 0.05 and 0.1 mg kg^{-1}) of okra and indicated as a relative standard deviation (RSD %) value. Intra-laboratory-based experimental reproducibility was checked using Horwitz ratio (HorRat)[19].

Data analysis

For residual quantification, the chromatographic peak area of the matrix-matched standard was compared to that of samples collected from the field experiment. Pesticide degradation kinetics was determined when the log residues (mg kg^{-1}) were plotted against time (days). The best-fit curve equation was determined from the maximum square of the coefficient of determination (R^2). Half-lives ($T_{1/2}$) and pre-harvest intervals (PHI) of pesticides were determined using the Hoskins[20] and Handa[21] formulae respectively.

Results and discussion

Method validation

Under optimized instrumental parameters dimethoate, acephate and methamidophos were eluted at 8.19, 5.20 and 3.71 min, with the best chromatographic separation in GC-FPD (Figure 1 *a*). In UHPLC, λ_{max} for emamectin benzoate was 254 nm, while for flubendiamide and des-iodo bendimide it was 230 nm. The corresponding retention time was 2.60, 6.29 and 8.49 min respectively (Figure 1 *b* and *c*). Based on the linearity with five different pesticide concentrations (x) against peak area (y), the calibration curves ($y = a + bx$) and a good coefficient of determination (R^2) of 0.99 were derived. The linear equation obtained from matrix-matched standards of okra fruit was nearly identical to the calibration curve of the standard in solvents (HPLC-grade hexane and acetonitrile). LOD was 0.003 mg kg^{-1}, while LOQ was 0.01 mg kg^{-1}. ME was within the prescribed limit of 3.63–7.63% (<20%) (Table 1). Recoveries were in the range 81.95–114.53% (Figure 1 *a–c*) and their intra-day precision RSD ranged from 1.78% to 5.00% (Table 2). The result complies with the criterion that the range of mean

Table 1. Linearity parameters and matrix effect of pesticides and their metabolite in/on okra fruit

Pesticide	Calibration range (mg l^{-1})	Linearity calibration curve ($y = a + bx$)		Coefficient of determination (R^2)		Matrix effect (%)
		Solvent	Okra matrix	Solvent	Okra matrix	
Dimethoate	0.01–1	$y = 2E + 07x - 32400$	$y = 2E+07x - 317796$	0.999	0.9993	5.90
Acephate	0.01–1	$y = 2E + 07x - 32855$	$y = 2E+07x - 207111$	0.999	0.9979	7.63
Methamidophos	0.01–1	$y = 1E + 07x - 27935$	$y = 1E+07x - 317567$	0.999	0.9986	3.63
Emamectin benzoate	0.01–0.1	$y = 83463x - 84.732$	$y = 88381x - 61.894$	0.9985	0.9987	6.62
Flubendiamide	0.01–0.1	$y = 31866x + 67.553$	$y = 35472x + 9.187$	0.9999	0.9976	5.76
Des-ido flubendimide	0.01–0.1	$y = 51492x + 134.36$	$y = 54453x + 140.64$	0.9996	0.9976	5.49

Table 2. Recovery level of pesticides and their metabolites in/on okra fruit

Pesticide	Spiking level (mg kg^{-1})								
	0.01			0.05			0.1		
	Recovery (%)* ± SD	RSD	HorRat	Recovery (%)* ± SD	RSD	HorRat	Recovery* (%) ± SD	RSD	HorRat
Dimethoate	94.11 ± 4.40	4.76	0.19	108.46 ± 4.02	3.71	0.19	114.53 ± 5.06	4.42	0.25
Acephate	95.84 ± 3.57	3.72	0.15	92.01 ± 3.87	4.20	0.21	97.14 ± 3.09	3.18	0.18
Methamidophos	98.86 ± 3.24	3.27	0.13	106.89 ± 4.38	4.10	0.21	102.06 ± 2.92	2.86	0.16
Emamectin benzoate	98.07 ± 1.87	1.91	0.08	95.82 ± 3.55	3.70	0.19	97.54 ± 3.39	3.47	0.20
Flubendiamide	90.09 ± 4.51	5.00	0.20	81.95 ± 1.68	2.05	0.10	87.00 ± 1.16	4.78	0.27
Des-ido flubendimide	96.63 ± 1.98	2.05	0.08	104.80 ± 1.87	1.78	0.09	103.06 ± 1.48	1.43	0.08

*Mean of three replicates; SD, Standard deviation; RSD, Relative standard deviation (intra-day); HorRat, Horwitz ratio.

recoveries at relevant concentrations is within 70–120%. In terms of repeatability, a satisfactory precision RSD ≤20% was obtained[17]. The reproducibility of methodology in terms of HorRat was within the prescribed range from <0.5 to 2 (Table 2)[22].

Dissipation pattern of pesticides in okra

Dimethoate: In okra, initially (1 h after spraying), dimethoate application at 300 g a.i. ha^{-1} deposited 3.39 mg kg^{-1} of residues (Figure 2 *a*). On 1, 3 and 5 days after treatment (DAT), the residues declined over time as 2.06, 0.85 and 0.23 mg kg^{-1} with 39.29%, 74.85% and 93.30% loss respectively. About 97% of the residues dissipated (0.08 mg kg^{-1}) on 7 DAT (Figure 2 *b*) and attained below the detectable limit at 10 DAT (BDL < LOQ) (Table 3). In okra and chilly, dimethoate (300 g a.i. ha^{-1}) application recorded a similar residue level of 2.93 and 3.12 mg kg^{-1} respectively, and about 90% and 80% of the residues dissipated on 10 and 5 DAT respectively[23,24]. On the contrary, a higher residue level of 8.01 mg kg^{-1} was initially deposited after dimethoate 30 EC (6 g a.i. ha^{-1}) application and residues were detected up to 21 DAT in okra[25]. This difference in dimethoate degradation might be because of the variation in plant species, spray interval, dosage, pesticide application method and other external environmental factors[26].

Acephate and methamidophos: In okra, acephate application at 292 g a.i. ha^{-1} deposited 3.21 mg kg^{-1} of residues initially (Table 3 and Figure 2 *c*). This declined over time as 2.55, 1.77, 1.27 and 0.51 mg kg^{-1} with 20.56%, 44.82%, 60.43% and 84.04% loss on 1, 3, 5 and 7 DAT respectively. More than 95% of acephate residues (0.10 mg kg^{-1}) dissipated on 10 DAT (Figure 2 *e*) and reached BDL on 15 DAT (Table 3). Initially (1 h of spraying), no methamidophos residue was detected. Acephate residues penetrated okra fruit and subsequently metabolized to methamidophos (0.06 mg kg^{-1}), detected on 1 DAT. Similarly, in capsicum and cucumber, though methamidophos was not formed on the same day after acephate application, it was detected on 1 DAT[27,28]. In the present study, about 2% of acephate residue was metabolized to methamidophos on 1 DAT, which increased to about 5.24% on 3 DAT (Figure 2 *d*). After this period, methamidophos residues quickly dissipated and reached BDL on 5 DAT. Faster dissipation of acephate metabolite or degradation to some other secondary metabolites was reported in the greenhouse and field-grown tomatoes[29]. In another study on okra, initially, 8.44 and 0.14 mg kg^{-1} of acephate and methamidophos residues respectively, were deposited after acephate application at 560 g a.i. ha^{-1}. Increased deposit and longer persistence (up to 15 DAT) of acephate might be due to a higher application dose (nearly twice the concentration of acephate in the present study). The dissipation trend of methamidophos was similar to the present study, with an increase in residue level up to 3 DAT followed by a decrease from 5 to 15 DAT[30].

Emamectin benzoate: In okra, emamenctin benzoate applied at 8.5 g a.i. ha^{-1} deposited 0.36 mg kg^{-1} of residues initially (Figure 3 *a*). About 82% of the residue dissipated on 3 DAT (Figure 3 *b*) and reached BDL rapidly on 5 DAT (Table 3). The results are in conformity with earlier findings

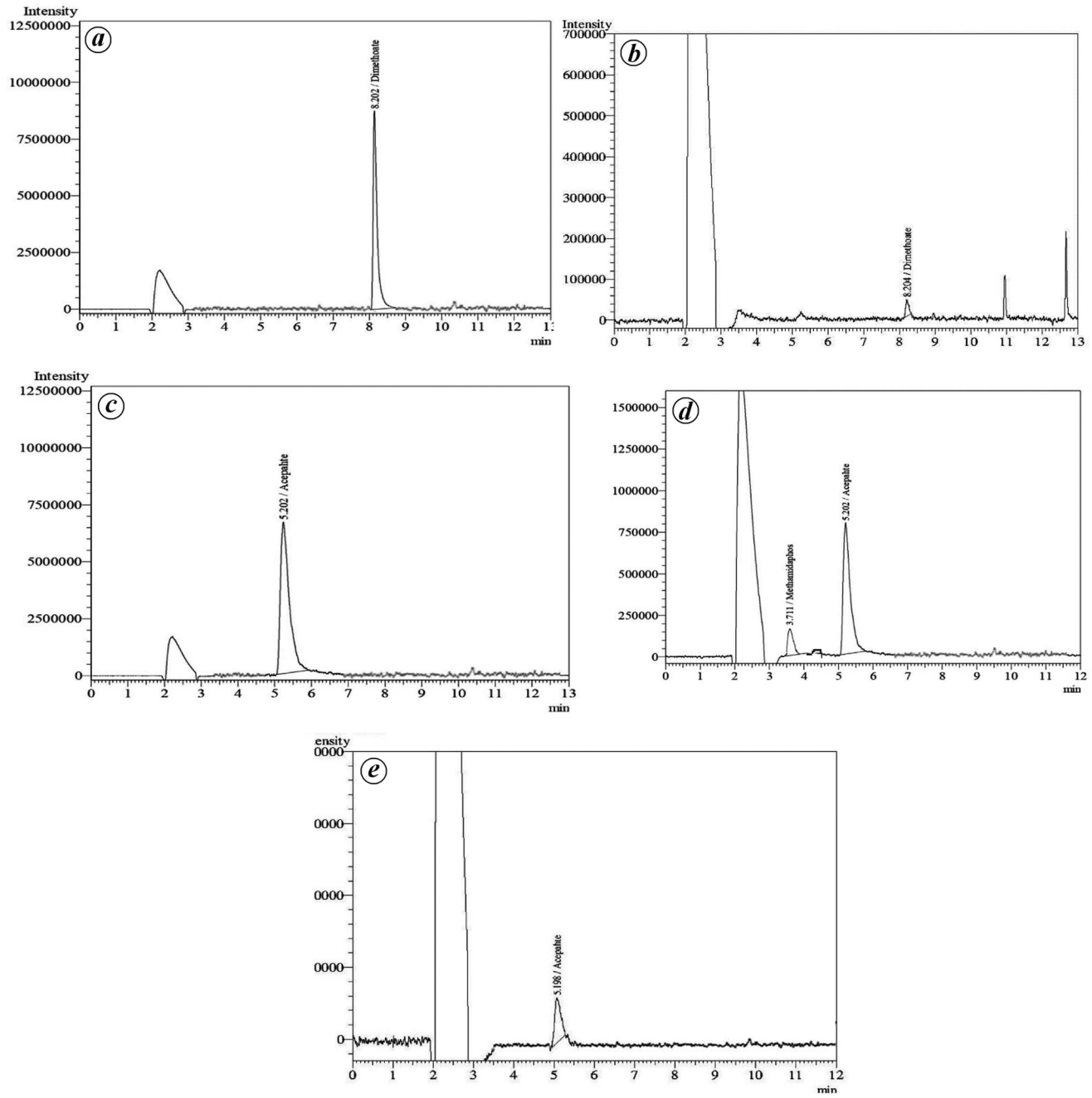

Figure 2. GC chromatogram of okra sample: (***a***) dimethoate – 0 days, (***b***) dimethoate – 7 DAT, (***c***) acephate residues – 0 days, (***d***) acephate and methamidophos residues – 3 DAT and (***e***) acephate residues – 10 DAT.

in okra with 0.302 mg kg^{-1} residue deposited initially after emamectin benzoate application (9 g a.i. ha^{-1}), which reached BDL on 5 DAT[31]. For three applications of emamectin benzoate in okra at a higher dose (68.1 and 136.2 g a.i. ha^{-1}), the residues dissipated quickly and reached BDL on 5 DAT for both doses[32]. In cabbage, around 55% of the applied emamectin benzoate (8.5 g a.i. ha^{-1}) residues dissipated on 1 DAT and reached BDL on 5 DAT[33]. Thus, it can be confirmed that emamectin benzoate has a faster dissipation rate in spite of differences in crop matrices.

Flubendiamide and des-ido flubendiamide: In okra, initially flubendiamide sprayed at 48 g a.i. ha^{-1} deposited 1.16 mg kg^{-1} of residue. On 1, 3 and 5 DAT, flubendiamide residue levels gradually declined to 0.54, 0.23 and 0.12 mg kg^{-1} with 53.63%, 79.68% and 90.06% loss respectively. Around 95% of the residues dissipated on 7 DAT (Figure 3 *d*) and reached BDL on 10 DAT (Table 3). In another study on okra, a similar pattern of flubendiamide (48 g a.i. ha^{-1}) dissipation was recorded, with around 86% of the initial residue (0.53 mg kg^{-1}) dissipated on 7 DAT

Table 3. Dissipation pattern of pesticide residues and their metabolites in/on okra fruit

	Residues (mg kg^{-1})											
	Dimethoate @ 300 g a.i. ha^{-1}		Acephate @ 292 g a.i. ha^{-1}		Methamidaphos		Emamectin benzoate @ 8.5 g a.i. ha^{-1}		Flubendiamide 480 SC @ 48 g a.i. ha^{-1}		Des-ido flubendiamide	
DAT	Mean* ± SD	Dissipation (%)	Mean* ± SD	Dissipation (%)	Mean* ± SD	Dissipation (%)	Mean* ± SD	Dissipation (%)	Mean* ± SD	Dissipation (%)	Mean* ± SD	Dissipation (%)
Control	ND	–	ND	–	ND	–	ND	–	ND	–	ND	–
0	3.39 ± 0.08	–	3.21 ± 0.11	–	BDL	–	0.36 ± 0.02	–	1.16 ± 0.02	–	BDL	–
1	2.06 ± 0.04	39.29	2.55 ± 0.04	20.56	0.06	–	0.24 ± 0.01	32.86	0.54 ± 0.02	53.63	BDL	–
3	0.85 ± 0.05	74.85	1.77 ± 0.03	44.82	0.10	–	0.06 ± 0.01	82.63	0.23 ± 0.01	79.68	BDL	–
5	0.23 ± 0.07	93.30	1.27 ± 0.05	60.43	0.06	–	BDL	100.00	0.12 ± 0.01	90.06	BDL	–
7	0.08 ± 0.01	97.77	0.51 ± 0.05	84.04	BDL	100.00	BDL	–	0.05 ± 0.01	95.62	BDL	–
10	BDL	100.00	0.10 ± 0.01	97.03	BDL	–	BDL	–	BDL	100.00	BDL	–
15	BDL	–	BDL	100.00	–	–	BDL	–	BDL	–	BDL	–

*Mean of three replicates; DAT, Days after treatment; ND, Not detected; BDL, Below detectable level (<0.01 mg kg^{-1}).

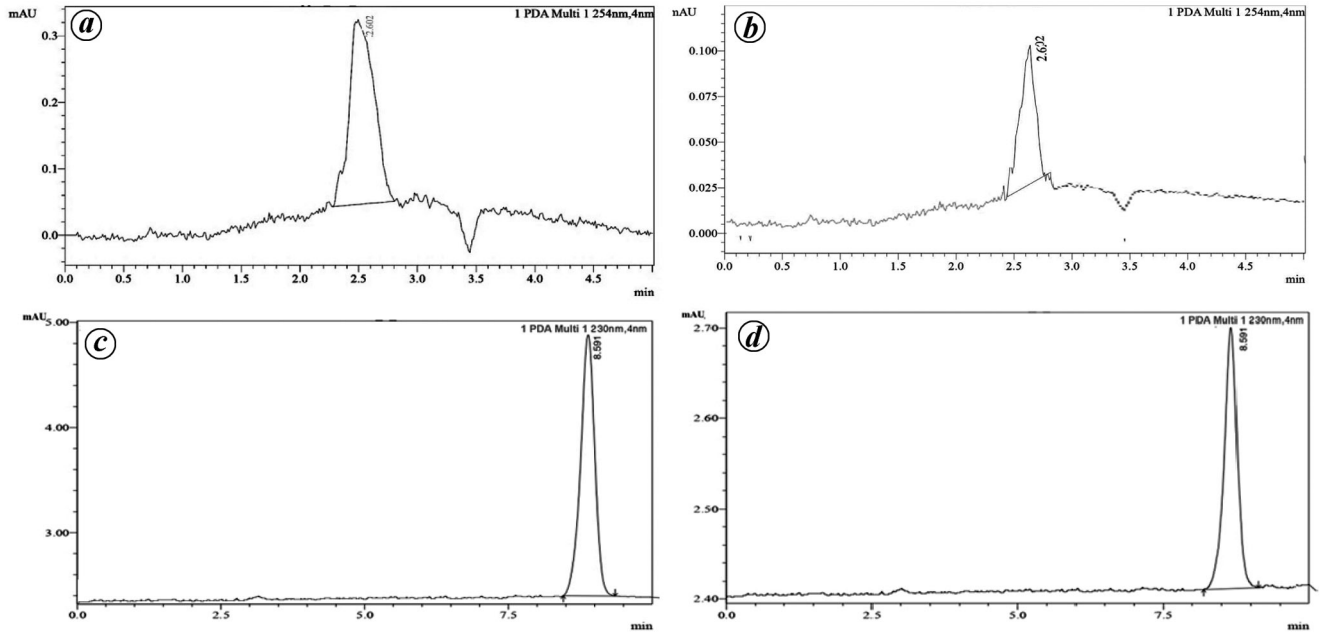

Figure 3. UHPLC chromatogram of okra sample: (***a***) emamectin benzoate – 0 days, (***b***) emamectin benzoate – 3 DAT, (***c***) flubendiamide – 0 days and (***d***) flubendiamide – 7 DAT.

and reached BDL on 10 DAT[34]. Likewise, in okra, 0.84 mg kg^{-1} of flubendiamide residues were deposited initially when applied at 60 g a.i. ha^{-1}. Around 98.80% of the residues dissipated on 7 DAT and reached BDL on 10 DAT[35]. During flubendiamide photo-degradation process, the formation of des-iodo flubendiamide (a metabolite) increases gradually up to a certain point and then declines regularly[36]. However, in the present study, throughout the sampling period, des-iodo flubendiamide was not detected (Table 3 and Figure 3 *c* and *d*). In many vegetable crops such as okra[34], cabbage[37], gherkin[38], tomato[39] and chillies[40], only flubendiamide residues were detected and not des-ido flubendiamide. This may be because of the low amount of des-ido flubendiamide formed during degradation.

Degradation kinetics of pesticides

When the residual log was plotted against time, a straight line was formed with a significant coefficient of determination (R^2 = 0.985–0.9954) (Table 4 and Figure 4). Thus, all the pesticides degraded through the first-order reaction pathway and followed the exponential equation

$$C_t = C_0 e^{-kt},$$

where C_0 is the apparent initial residue level (mg kg^{-1}), C_t the pesticide residue level at time t (mg kg^{-1}) and k is the degradation rate constant.

Table 4. Degradation kinetics of dimethoate, acephate, emamectin benzoate and flubendiamide in/on okra

Pesticide	Regression equation ($y = a + bx$)	Coefficient of determination (R^2)	First-order equation	k-value	Half-life (days)	MRL (mg kg^{-1}) for okra		PHI (days)	
						FSSAI	EU	FSSAI	EU
Dimethoate	$y = 3.5622–0.2376x$	0.9954	$y = 3.6491e^{-0.547x}$	0.547	1.27	2 (vegetables)	0.01	1	10.65
Acephate	$y = 3.5276–0.1037x$	0.9881	$y = 3.3697e^{-0.239x}$	0.239	2.90	Not fixed	0.01	–	24.5
Emamectin benzoate	$y = 2.5914–0.2592x$	0.9869	$y = 0.3903e^{-0.597x}$	0.597	0.50	0.05	0.02	3.32	4.86
Flubendiamide	$y = 2.9816–0.1858x$	0.9874	$y = 0.9479e^{-0.419x}$	0.419	1.65	Not fixed	0.01	–	11.33

MRL, Maximum residual limit; FSSAI, Food Safety Authority of India; EU, European Union; PHI, Pre-harvest interval.

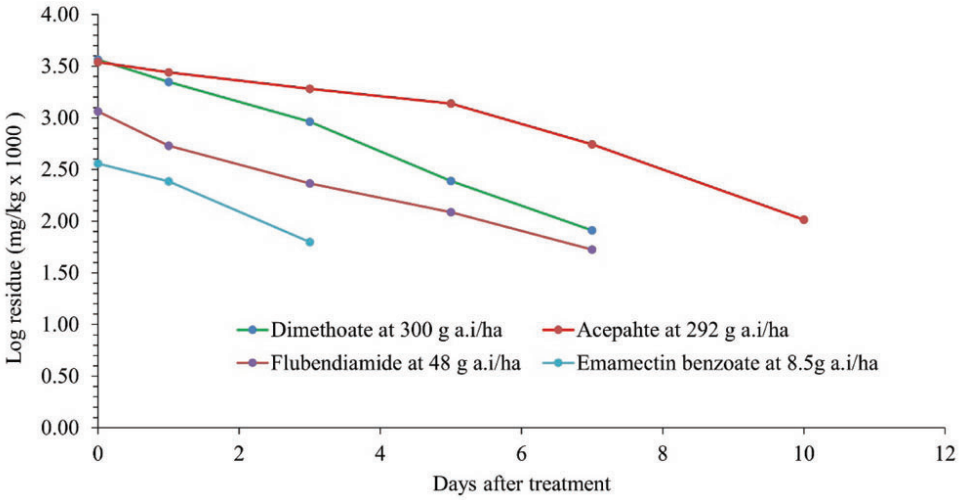

Figure 4. Semi-logarithmic graph showing degradation kinetics of pesticides in/on okra.

Acephate took the longest time of 2.90 days for degradation to half of its level deposited initially ($T_{1/2}$), followed by flubendimide (1.65 days), dimethoate (1.27 days) and emamectin benzoate (0.5 days). In okra, $T_{1/2}$ of 2.70 and 3.0 days was recorded after acephate (560 and 1120 g a.i. ha^{-1}) application, which was in close agreement with the present study[30]. $T_{1/2}$ of flubendiamide in the present study was similar to the findings in tomato (50 g a.i. ha^{-1})[36]. Though a higher dimethoate $T_{1/2}$ (4.2 and 5.21 days) has been reported in okra[25,41], the present value (1.27 days) is in close proximity with those reported in chilli, 1.94 days[42] and 1.74 days[24]. When compared to other pesticides under study, $T_{1/2}$ value of emamectin benzoate (0.5 days) was shorter. Similarly, another study on okra had a shorter $T_{1/2}$ of 0.6 days for emamectin benzoate (12 g a.i. ha^{-1})[43]. Likewise, in cabbage, a shorter $T_{1/2}$ of 1.34–1.72 days[44] and 1–1.1 days[45] was recorded during the rapid emamectin benzoate (9 and 18 g a.i. ha^{-1} respectively) dissipation process. Based on European Union (EU) MRL[46], the highest PHI of 24.5 days was arrived for acephate, followed by 11.77, 10.65 and 4.86 days for flubendiamide, dimethoate and emamectin benzoate respectively. Based on the available FSSAI MRL for dimethoate and emamectin benzoate[47], PHI was 1 and 3.32 days respectively (Table 4). Similar to the present study, a higher PHI of 25 days for acephate in okra was reported[30]. For dimethoate, PHI of 13.63 days[41] and 9.38 days[24] in chilly and 18.35 days in okra[25] have been recorded.

Conclusion

Based on the dissipation pattern, average initial deposits of dimethoate and acephate (3.39 and 3.21 mg kg^{-1} respectively) were comparatively higher than flubendiamide and emamectin benzoate (1.16 and 0.36 mg kg^{-1} respectively). Rapid residue dissipation was observed for emamectin benzoate residue (3 DAT), while dimethoate (7 DAT), flubendiamide (7 DAT) and acephate (10 DAT) showed prolonged dissipation. Acephate residues metabolized to methamidophos were detected on 1 DAT and reached a maximum on 3 DAT. Des-ido flubendiamide residues were not detected throughout the sampling period. All the pesticides degraded through first-order reaction with $T_{1/2}$ of 1, 27, 2.90, 0.5 and 1.65 days for dimethoate, acephate, emamectin benzoate and flubendiamide respectively. Acephate residues persisted much longer, nearly twice the time compared to other pesticides. After two sprayings of dimethoate, emamectin benzoate and flubendiamide, following PHI (EU MRL) of 3–11 days is mandatory for the production of residue-free okra. Okra being harvested on alternate days,

following PHI of 24 days after acephate application, is not possible. Thus, the use of acephate at the flowering stage poses the risk of residue detection in harvested okra. Educating the farmers about pesticide recommendations for a crop, their dosage against target insect pests and PHI are the ways to harvest okra free from pesticides.

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