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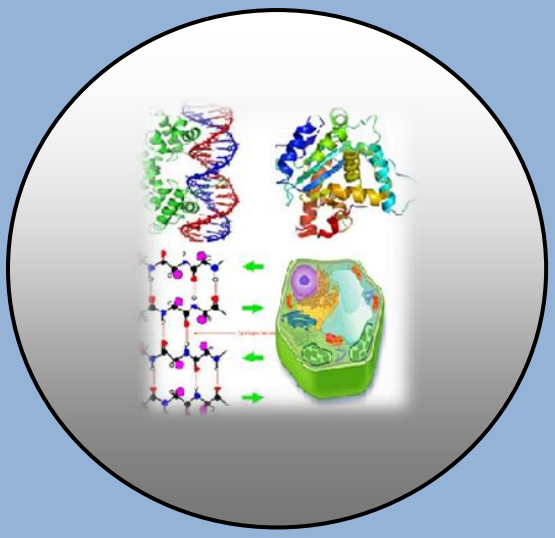

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Physicochemical Properties of Ten Commercial Chlorpyrifos Emulsifiable Concentrate Formulations and their Biological Effects on Pink Bollworm *(Pectinophora gossypiella)*

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ABSTRACT

The present study was conducted to investigate the effect of different storage conditions on ten commercial formulations of chlorpyrifos (48% w/v) emulsifiable concentrates in the Egyptian market. The formation of sulfotep (relevant impurity of chlorpyrifos), pH, conductivity, refractive index, density, effect of centrifugation, persistent foam and emulsion stability were determined. The insecticidal activity of the tested formulations on pink bollworm Pectinophora gossypiella was also evaluated. The results showed that chlorpyrifos content and amount of sulfotep in the tested different formulations in the acceptance range according to FAO specifications. Also, all tested formulations had acidic character. Results also indicated that all chlorpyrifos formulation passed successfully through emulsion stability and re-emulsification test before and after storage at 0 °C and 54 °C for 7 and 14 days, respectively and four freeze-thaw cycles when the formulations diluted with CIPAC standard water A and D except source 10 in all storage conditions and source 5 and 6 in freeze thaw cycles in CIPAC standard waters D and A, respectively. No phase separation or sediment observed in all formulations after centrifugation and the volume of foam from the different formulations was low and passed through the recommended rate of foam. The Source 1 was not detected in its content of sulfotep. Meanwhile, the source 5 was the highest in sulfotep content. The most effective formulation against Pectinophora gossypiella was source 8 had the LC_{50} value 290.73 ppm and the lowest effective formulation was source 10 recorded LC_{50} value 1647.56 ppm.
Keywords: Chlorpyrifos, Sulfotep, Physical Properties and Pectinophora gossypiella.

INTRODUCTION

Chlorpyrifos (CPF) is one of the most widely used active ingredients in organophosphorus insecticides in agriculture and non-agriculture applications (Farahat et al., 2011; Sasikala et al., 2012; Ramzy, et al., 2014; Abdelmonem, 2015; Diqiu et al., 2015). The important agricultural crops such as soybeans, wheat, alfalfa, citrus, tree nuts, peanuts, vegetables, and others, from yield caused by insect pests (Pope et al., 2005). WHO (1997) classified chlorpyrifos as a moderately dangerous, Class II insecticide. Products containing chlorpyrifos have been on the market for more than 40 years (DAS, 2009).Today, chlorpyrifos is registered in more than 98 countries worldwide for use on more than 50 different crops against damage caused by a wide range of insect pests. Sulfotep is a highly toxic impurity that may be present in trace quantities in chlorpyrifos. Sulfotep is relatively a stable toxic impurity that may concentrate in the environment and causing unanticipated health and ecological problems, so that the level of sulfotep is limited to be 0.3% as the maximum concentration in chlorpyrifos formulations FAO (2008). The pink bollworm *Pectinophora gossypiella* (Saunders) is one of the most injurious cotton pests in the world (Lykouressis et al. 2005; Al-kazafy et al., 2014; Ezzat et al., 2015). It is found in almost every cotton-producing country and has caused a lot of damage. Protection of cotton plants and mass production of harvested cotton fibres depends mainly upon the efficient control of the *Pectinophora gossypiella*. Thus, the objective of this study was to investigate the effect of different storage conditions on the stability of ten commercial chlorpyrifos (48%) emulsifiable concentrate (EC) formulations (manufactured from various companies), presence of its impurity; sulfotep and physical properties and insecticidal activity of the tested formulations on pink bollworm.

MATERIAL AND METHODS

Chemicals

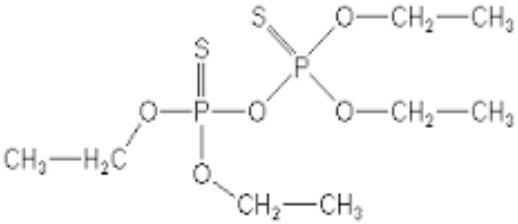
Calcium carbonate was purchased from Sigma- Aldrich Chemie GmbH Steinheim, Germany, Magnesium oxide and methyl red were purchased from Qualikems Fine Chemicals. India. Ammonia Solution and methanol were purchased from Prolabo. Water used obtained from Water distiller LABCONCO water PROT.M PS LABCONCO Corporation, Kansas City, Missouri 64132-USA. Chlorpyrifos: analytical standard 99.4% from AAKO company The Netherlands. Sulfotep analytical standard 97.1%: from Chem. Service. Chlorpyrifos 48% (EC) commercial formulation was obtained from ten different manufacturers in Egypt.

IUPAC of Chlorpyrifos: O, O-diethyl O-(3, 5, 6-trichloro-2-pyridyl) phosphorothioate.

Structural formula

IUPAC of sulfotep: (*O,O,O',O'*-tetraethyl dithiopyrophosphate)

Structural formula



Test Insect

Field populations of *P.gossypiella* were collected from Sharkia governorate during 2014 cotton season. The infested green bolls were collected at the end of the cotton growth season; the disposed larvae were released from the double infested seeds and reared under constant temperature 27 ± 2°C and 70 ± 5% relative humidity (Rashad and Ammar, 1985).

Preparation of Samples

Preparation of chlorpyrifos standard

Ten mg of chlorpyrifos analytical standard was weighed inside a 25 milliliter volumetric flask then dissolved and completed to the final volume with methanol.

Sample preparation for tested chlorpyrifos

Accurately weight sufficient sample material formulation 48% w/v to equivalent ten mg of chlorpyrifos standard in a 25 milliliter volumetric flask for each sample and slowly mixed with methanol and complete the volume with methanol.

Preparation of sulfotep standard

Ten mg of sulfotep analytical standard was weighed inside a 10 milliliter volumetric flask then dissolved and completed to the final volume with methanol.

Sample preparation of sulfotep impurity

One gram of all tested formulation samples (chlorpyrifos 48% w/v) was weighed containing 0.48 g in different volumetric flask dissolved with methanol and completed to the final volume with methanol.

Determination of chlorpyrifos and sulfotep impurities by Gas chromatography

Chlorpyrifos and sulfotep were determined according to the method of DAS (2004) with some modification using GLC. A Hewlett-Packard 6890 gas chromatograph equipped with Flame Ionization Detector (FID) at 275°C, capillary column HP-50% (15 m x 0.53 mm I.D., 1 µm film thickness). Nitrogen was used as a carrier gas at 7 ml/min. The oven temperature program was held at 160 °C for 1 min, then ramp 10°C / min to 250°C and kept at that temperature for 5 min. Injector temperature 200°C. The injection volume was 1 µl. Chlorpyrifos and sulfotep were quantitively determined by comparison with the standard of known purity under the identical GLC conditions.

Physical properties of chlorpyrifos 48% EC formulations

Storage stability

The accelerated storage tests (storage stability) were carried out according to CIPAC methods [CIPAC MT 39.3 and CIPAC MT 46.3]. The storage (stability) test (0°C) was performed during one week and the storage test (54°C) during two weeks.

Freeze -Thaw Cycles

Freeze-thaw cycles are a method of putting stress on the formulation to stimulate the conditions that are encountered in warehouse storage. Test tubes filled with the prepared formulation and hermitically closed were vertically stored for 12h in freezer at -20°C, and then for 12h at room temperature 25°C±2. The formulations were observed for any change recorded. The formulation is considered "stable" if there is no substantial separation after four cycles.

Centrifugation test

The centrifugation is a relatively simple method and allows one to accumulate a large set of data for a relatively short period of time. Formulations were subjecting to centrifugation at speeds up to 5400 rpm for 5 min by using a Laboratory Centrifuge REMI Centrifuge REMI Equipments Bombay-India- R32A.4000002.The formulation was centrifuged at 25°C.

Persistent foam

Persistent foam is a measure for the amount of foams likely to be present in a spray tank or other application equipment following dilution of the product with water. Specified amount of the material is added to CIPAC standard water (95ml) in the measuring cylinder and made up to the mark. The cylinder is stoppered and inverted 30 times. Stand the cylinder on the bench and left undisturbed for the specified time. The volume of foam was noted [CIPAC MT 47.2].

pH Measurement

pH value of chlorpyrifos 48% EC formulations was measured by using a pH Meter "Jenway Instruments pH 3510 pH meter. It was recalibrated before testing. [CIPAC MT 75.3].

Conductivity Measurement

The conductivity of the different formulations was measured by Conductivity and Salinity meter "Thermo Orion model 115A$^+$, USA". The measurements were made at 25°C ±1. Before the measurement, the conductometer was calibrated with 0.01M KCl solution at temperature (25°C ±2).

Refractive index

Refractive index is an optical measurement of a materials ability to bend a beam of light; the refractive index could be used to determine the purity of the material. Refractive index of the different formulations was measured by using ABBE Refractometer, ATAGO, Co., LTD, Japan (ASTM, 2002).

Density measurement

Density of the different formulations was measured using digital density meter model DDM 2910 by touch screen. Rudolph Research Analytical, USA.

Viscosity measurement

Viscosity of the different formulations was measured without dilution, using Brookfield DV II$^+$ PRO digital Viscometer. (Brookfield, USA). UL rotational adaptor. The temperature was kept at 25°C during the measurement by water bath TC-502. USA and each reading was taken after equilibrium of the sample (ASTM, 2010).

Emulsion stability and re-emulsification (MT 36.3)

The formulation, when diluted at 30 ± 2°C with CIPAC Standard Waters A and D. (MT 18).In the emulsion characteristics experiment, 5 ml of the formulation samples were separately mixed with standard water (CIPAC A, 20 ppm hardness, pH 5.00-6.00, Ca^{2+}:Mg^{2+}=1:1 and CIPAC D, 342 ppm hardness, pH 6.00-7.00, Ca^{2+}: Mg^{2+} = 4:1) in a 100 ml measuring cylinder to produce 100 ml of aqueous emulsion. The stopper was placed on the cylinder, which was subsequently turned upside down 10 times. Subsequently, the amount of free oil or cream that separated at the top or the bottom of the emulsion was observed after the emulsion was allowed to stand undisturbed for various intervals (0, 0.5, 2, 24 h and 24.5). For the stability test at low temperature (0°C), 100 ml of each sample was transferred to a glass tube. For cooling, the tube and its contents were placed in a refrigerator and remained at 0°C for 7 days. At the end of 7 days, the tube was removed from the refrigerator, and allowed to remain undisturbed at room temperature for 3 h. The volume of any separated material at the bottom of the tube was subsequently recorded. Accelerated storage procedure was executed by placing the samples (about 50 ml each) in bottles and placing the capped bottles and contents in an oven of 54°C for 14 days.

Toxicity of the tested different formulations against *Pectinophora gossypiella*

Five concentrations were used for each formulation and three replicates of ten adults were used for each concentration. Glass chimney cages (6 x 9cm) were dipped in water dilution of each formulation for 20 seconds and left to dray. The newly moth (zero day old) were exposed to residual tested formulations in glass chimney cages and covered with muslin cloth to allow air circulation. Percentage mortality was calculated after twenty-four hours of exposure and corrected by Abbott's formula (1952). The slop and LC_{50} values for each formulation were calculated according to Finney (1971). Also the toxicity index calculated according to Sun (1950).

RESULTS AND DISCUSSION

Effect of different storage conditions on the content of chlorpyrifos 48% emulsifiable concentrate.

Data in Table 1 showed the degradation of the commercial formulations of chlorpyrifos 48% (EC) obtained from ten different manufacturers in Egypt (manufactured from ten different companies) was in the acceptance range of specification of FAO (2008), the content of chlorpyrifos after storage at 54°C for 14 days should not be lower than 95% relative to the content of chlorpyrifos before storage but the active ingredient in the tested chlorpyrifos formulations were affected by storage conditions and period of exposure. The temperature at 54°C was effective in the decomposition percentage of active ingredient chlorpyrifos more than 0°C and four freeze thaw cycles. This is agreement with many authors (Rahman and Motogoua, 2000; Wu et al., 2006; Stenrod et al., 2008) reporting the degradation of chlorpyrifos was slower at low temperature and was markedly stimulated by increasing the temperature. Also, NAR (2000) reported that the chlorpyrifos is thermally sensitive to temperature over 50°C and breakdown relatively quickly in the environment.

Effect of different storage conditions on the formation of sulfotep.

Sulfotep is the main impurity in chlorpyrifos (technical and formulations), it should not be higher than 3g/kg in all formulations of chlorpyrifos (FAO, 2008). Sulfotep is a highly toxic impurity that may be present in trace quantities in chlorpyrifos Ambrus et al., (2003). The data presented in Table 2 demonstrated that source (1) was not detected in its content of sulfotep, while all remaining tested formulations were moderate in their content of sulfotep except source (5) as the highest in sulfotep content, but the amount of sulfotep in the all formulations of chlorpyrifos in acceptance range of FAO specifications and evaluations for chlorpyrifos (2008). The level of sulfotep is limited to be (3 g/kg or 0.3%).This indicate that the amount of sulfotep present in the commercial formulations varies from one sample to another, and that depend on the manufacturing practice, the storage conditions and inert ingredients used for formulation pesticides such as solvents, surfactants. These results are in harmony with those obtained by Fakhraian et al., (2004) stated that the formation of sulfotep (the major impurity) during the synthesis of chlorpyrifos is influenced by the nature and concentration of the catalyst, temperature, stirring and time of reaction and also these obtained results were agreement with Allender and James (1991) they reported that the sulfotep content of the commercial products did not show any correlation with storage time.

Physical properties different formulation of chlorpyrifos 48% EC

All the tested formulations exhibited acidic pH value. The pH values were in range (4.82-5.64) in the acceptance limit of FAO specification. The formulations having acidic character implying that they will have good biological activity (Molin and Hirase, 2004). The tested formulations having conductivity range (0.1-0.3). The variation of density was 1.068-1.091 g/cm^3. The viscosity data of these ECs were found in the range of (2.36-3.95 mPas) at 100 rpm. The volume of foam from the different formulations is low and passed through the recommended rate of foam (Maximum: 20 ml after 1 minute).

Effect of different storage conditions on the emulsion stability and reemulsification

The data in Table 7-10 showed the emulsion stability and reemulsification of the ten commercial chlorpyrifos formulations (manufactured from various companies) after storage at 0°C and 54 °C for 7 and 14 days, respectively. The formulation, when diluted at 30 ±2 °C with CIPAC standard waters A and D shall comply with the specifications of chlorpyrifos emulsifiable concentrate. Results indicated that all chlorpyrifos formulations passed successfully through emulsion stability and re-emulsification test before and after storage at 0°C and 54°C for 7 and 14 days and four freeze-thaw cycles when formulation diluted with CIPAC standard water A and D except source 10 in all storage conditions and source 5 and 6 in freeze thaw cycles in CIPAC standard waters D and A, respectively. According to JMPS FAO/WHO pesticides specifications, 2010; the maximum level of cream and precipitate layer should don't exceed about 2 ml after 0.5, 2, and 24.5 hrs from dilution.

Biological activity of the tested different formulations against *Pectinophora gossypiella*

The LC_{50} values are tabulated in Table 11 with their corresponding slopes and toxicity index. The results showed the efficiency of the tested formulations against *Pectinophora gossypiella.* Source 8 formulation was the most effective at the LC_{50} level, whereas the source 10 formulation was the last effective. The tested ten formulations could be classified into three categories at LC_{50} level. The first category include source 8 and 4 which gave the highest effect against *Pectinophora gossypiella*. The LC_{50} values were 290.73 and 326.6 ppm, respectively. The second category included source 6, 9, 5 and 2 which had the LC_{50} values 786.6, 967.39, 967.39 and 1009.7 ppm, respectively. The third category include the source 3, 1,7 and 10 which had LC_{50} values 1274.8, 1440.02, 1561.93 and 1647.56 ppm, respectively. The toxicity index by comparing the toxicity of tested formulations, at a fixed level LC_{50} to their most effective compound. Since source 8 was most toxic formulation among the tested ones, it was used as a standard in calculating the toxicity index at LC_{50} level. Finally the data showed slope value ranging between (0.66 -2.520).

CONCLUSION

The emulsifiable concentrate formulations of the chlorpyrifos were characterized based on active ingredient content, sulfotep content, pH, conductivity, refractive index, viscosity, density, effect of centrifugation, persistent foam, emulsion stability and biological activity. The results showed that the chlorpyrifos content and amount of sulfotep in the ten different formulations were in the acceptance range and all tested formulations having acidic nature. Results indicated that all chlorpyrifos formulation passed successfully through emulsion stability and re-emulsification test before and after storage at 0°C and 54°C for 7 and 14 days and four freeze-thaw cycles when formulation diluted with CIPAC standard water A and D except source 10 in all storage conditions and source 5 and 6 in freeze thaw cycles in CIPAC standard waters D and A, respectively.

The volume of foam from the different formulations is low and passed through the recommended rate of foam. The Source 1 was not detected in its content of sulfotep. Meanwhile, the source 5 was the highest in sulfotep content. The most effective formulation against *Pectinophora gossypiella* was source 8 had the LC_{50} values 290.73 ppm and the toxicity index was 100 and the lowest effective formulation was source 10 recorded LC_{50} value 1647.56 ppm and lowest toxicity index 17.64 .Source 10 was the last effective, this may be due to failed in the emulsion stability and reemulsification when diluted with CIPAC standard water A and D.

Table 1. Effect of different storage conditions on the content of chlorpyrifos 48 % EC formulations from ten different manufacturers.

Formulations no.	Initial formulation room temperature	7 days 0°C		14 days 54°C		Freeze-thaw 4 cycles	
	chlorpyrifos content %	chlorpyrifos %	% Loss	chlorpyrifos %	% Loss	chlorpyrifos %	% Loss
1	47.90	47.68	0.46	47.07	1.76	47.38	1.09
2	47.66	47.36	0.63	46.83	1.77	47.40	0.55
3	47.71	47.44	0.57	46.49	2.62	47.28	0.91
4	47.61	47.47	0.29	47.08	1.13	47.17	0.93
5	47.75	47.28	0.99	46.79	2.05	47.16	1.25
6	47.79	47.27	1.1	46.32	3.17	46.81	2.09
7	47.73	47.43	0.63	47.02	1.51	47.29	0.93
8	47.92	47.75	0.63	47.13	1.68	47.22	1.48
9	47.86	47.39	0.99	47.09	1.64	47.12	1.57
10	47.63	47.42	0.44	46.93	1.49	47.05	1.23

Table 2.Effect of different storage conditions on the amount of sulfotep in chlorpyrifos 48 % EC formulations from ten different manufacturers.

F. No.	Initial temp.			0°C			54°C			Freeze-thaw cycles		
	chlorpyrifos content %	% sulfotep	Sulfotep as g/kg	chlorpyrifos content %	% sulfotep	Sulfotep as g/kg	chlorpyrifos content %	% sulfotep	Sulfotep as g/kg	chlorpyrifos content %	% sulfotep	Sulfotep as g/kg
1	47.90	UND	-	47.68	UND	-	47.07	UND	-	47.38	UND	-
2	47.66	0.013	0.273	47.36	0.011	0.232	46.83	0.016	0.342	47.40	0.007	0.15
3	47.71	0.006	0.14	47.44	0.012	0.253	46.49	0.0079	0.17	47.28	0.004	0.09
4	47.61	0.038	0.798	47.47	0.032	0.67	47.08	0.029	0.62	47.17	0.030	0.64
5	47.75	0.073	1.53	47.28	0.106	2.24	46.79	0.104	2.22	47.16	0.096	2.04
6	47.79	0.029	0.61	47.27	0.025	0.53	46.32	0.0113	0.244	46.81	0.0079	1.069
7	47.73	0.038	0.79	47.43	0.032	0.067	47.02	0.033	0.71	47.29	0.030	0.65
8	47.92	0.062	1.29	47.75	0.052	1.09	47.13	0.053	1.12	47.22	0.049	1.04
9	47.86	0.0253	0.53	47.39	0.025	0.527	47.09	0.021	0.45	47.12	0.022	0.47
10	47.63	0.072	1.51	47.42	0.078	1.64	46.93	0.069	1.47	47.05	0.068	1.44

Table 3.Physical properties of chlorpyrifos 48% EC formulations of ten different manufacturers at initial time.

Physical Properties	1	2	3	4	5	6	7	8	9	10
pH value (1%)	5.30	5.34	4.82	4.89	5.44	5.61	5.63	5.44	5.41	5.50
Conductivity	0.1	0.2	0.2	0.1	0.1	0.1	0.1	0.1	0.3	0.1
Refractive index	1.510	1.509	1.510	1.514	1.512	1.513	1.519	1.5063	1.515	1.516
Density(g/cm^3)	1.077	1.074	1.081	1.074	1.068	1.076	1.074	1.069	1.089	1.082
Viscosity (mpas)	3.41	3.71	3.68	3.56	2.36	2.53	2.53	2.98	3.80	3.91
Persistence foam (cm^3)	1	3	-	5	5	3	2	5	10	10

Table 4.Physical properties of chlorpyrifos 48% EC formulations of ten different manufacturers at 0°C for 7 days.

Physical Properties	1	2	3	4	5	6	7	8	9	10
pH value (1%)	5.29	5.40	5.70	4.87	5.22	5.32	5.38	5.32	5.18	5.37
Conductivity	0.1	0.2	0.2	0.2	0.1	0.1	0.1	0.1	0.3	0.1
Refractive index	1.510	1.5098	1.5098	1.5148	1.5127	1.5095	1.5087	1.506	1.5155	1.5155
Density(g/cm3)	1.077	1.074	1.082	1.074	1.069	1.074	1.073	1.069	1.089	1.062
Viscosity (mpas)	3.38	3.56	3.68	3.59	2.40	2.54	2.53	3.01	3.93	2.94
Persistence foam (cm^3)	1	10	1	10	5	5	10	5	10	10

Table 5. Physical properties of chlorpyrifos 48% EC formulations of ten different manufacturers at 54°C for 14 days.

Physical Properties	1	2	3	4	5	6	7	8	9	10
pH value (1%)	5.31	5.33	4.91	4.97	5.29	5.34	5.31	5.40	5.33	5.46
Conductivity	0.2	0.2	0.1	0.2	0.2	0.1	0.1	0.2	0.3	0.1
Refractive index	1.5014	1.5013	1.5125	1.5156	1.5133	1.5151	1.5138	1.5062	1.5159	1.5178
Density(g/cm^3)	1.080	1.078	1.095	1.081	1.075	1.091	1.079	1.069	1.091	1.083
Viscosity (mpas)	3.44	3.72	3.77	3.56	2.41	2.58	2.56	3.52	3.95	3.96
Persistence foam (cm^3)	2	10	2	10	10	10	8	10	10	10

Table 6. Physical properties of chlorpyrifos 48% EC formulations of ten different manufacturers at four freeze thaw cycles.

Physical Properties	1	2	3	4	5	6	7	8	9	10
pH value (1%)	5.34	5.38	4.91	4.95	5.30	5.42	5.40	5.5	5.62	5.64
Conductivity	0.1	0.5	0.1	0.1	0.1	0.1	0.1	0.1	0.3	0.1
Refractive index	1.5107	1.5106	1.5110	1.515	1.5138	1.5141	1.514	1.5107	1.5153	1.5164
Density(g/cm^3)	1.077	1.074	1.081	1.074	1.068	1.078	1.074	1.069	1.090	1.082
Viscosity (mpas)	3.46	3.74	3.79	3.58	2.43	2.59	2.56	3.57	3.93	3.95
Persistence foam (cm^3)	3	10	3	10	7	5	5	10	5	5

Table 7. Emulsion stability and reemulsification of ten commercial chlorpyrifos formulations 48 % EC at initial temperature.

F.no	Emulsion Characteristics											
	CIPAC A(CL*)						CIPAC D (CL*)					
	0.5h	1h	2h	4h	24h	24.5	0.5h	1h	2h	4h	24h	24.5h
1	2	2	2	2	2	-	1	1	1	1.5	2	1
2	-	-	Trace	Trace	Trace	-	-	-	-	-	0.1	-
3	0.1	0.1	1	1	1	0.1	-	-	-	-	0.1	-
4	-	-	1	0.5	1	0.1	-	-	-	-	-	-
5	1.5	2	2	2	2	1.5	Trace	0.1	0.1	0.2	1	Trace
6	1	1	1.5	3	3	1	-	-	-	-	0.1	-
7	1	1	2	3	3	1	-	-	-	-	0.1	-
8	-	-	-	1	1	-	-	-	-	-	-	-
9	-	-	-	Trace	Trace	-	-	-	-	-	-	-
10	2	3	3	5	5	2.2	2	2	2	2	2	3

***CL: Creamy layer**

Table 8.Emulsion stability and reemulsification of ten commercial chlorpyrifos formulations 48 % EC at 0 °C for 7 days.

F.no	Emulsion Characteristics											
	CIPAC A(CL*)						CIPAC D (CL*)					
	0.5h	1h	2h	4h	24h	24.5	0.5h	1h	2h	4h	24h	24.5h
1	1	1.8	2	2	2	1	0.5	0.5	1	1	2	0.5
2	-	-	-	-	-	-	-	-	-	-	Trace	-
3	-	0.1	0.5	1	1	-	-	-	-	-	-	-
4	-	-	0.1	0.5	1	-	-	-	-	-	2	-
5	0.5	1	1.5	2	2	2	-	0.1	0.2	0.2	2	-
6	0.5	1	1.5	2	3	0.5	-	-	-	Trace	0.5	-
7	0.5	1	1.5	2	2	0.5	-	-	-	-	0.5	-
8	-	-	0.1	1	1	-	-	-	-	-	Trace	-
9	-	-	-	-	0.5	-	-	-	-	-	-	-
10	1	2	3	3	4	2	1	1	2	2	3	1

***CL: Creamy layer**

Table 9. Emulsion stability and reemulsification of ten commercial chlorpyrifos formulations 48 % at 54 °C for 14 days.

F.no	Emulsion Characteristics											
	CIPAC A(CL*)						CIPAC D (CL*)					
	0.5h	1h	2h	4h	24h	24.5	0.5h	1h	2h	4h	24h	24.5h
1	1	1	1	1	2	1	-	1	2	2	2	1
2	-	-	-	-	-	-	-	-	-	-	0.1	-
3	-	-	-	-	-	Trace	-	-	Trace	0.2	0.3	-
4	1	1	1	1	1	-	-	-	Trace	0.1	1	1
5	0.5	0.5	1	2	2	1	1	1	2	2	2.5	1
6	0.5	0.5	1	1	2	1	0.5	0.5	1	1	2	0.5
7	-	-	-	1	2	1	0.2	0.5	1	1	2	0.5
8							-	-	-	Trace	0.5	-
9	-	-	-	0.5	0.5	-	-	Trace	0.5	1	2	-
10	-	-	-	2	3	1	2	2	3	3	4	2

***CL: Creamy layer**

Table 10. Emulsion stability and reemulsification of ten commercial chlorpyrifos formulations 48 % EC at four Freeze thaw cycles.

F.no	Emulsion Characteristics											
	CIPAC A(CL*)						CIPAC D (CL*)					
	0.5h	1h	2h	4h	24h	24.5	0.5h	1h	2h	4h	24h	24.5h
1	0.5	0.5	1	1.5	2	1	1	2	2	2	2	1
2	-	-	-	-	-	-	-	-	-	-	0.2	-
3	-	-	-	-	-	-	-	-	0.5	0.5	1	Trace
4	-	-	-	-	-	-	-	-	-	0.1	1	-
5	-	-	Trace	1	1	-	1	2	2	2	5	1
6	0.5	1	2	-	5	0.5	0.5	1	1	1	2	0.5
7	-	-	-	-	Trace	-	0.5	1	1	1	2	0.5
8	-	-	-	-	Trace	-	-	-	-	-	1	2
9	-	-	-	-	-	-	-	-	-	-	0.5	-
10	1	2	2	3	3	-	1	3	3	3	5	1

***CL: Creamy layer**

Table 11.Toxicity of different chlorpyrifos formulations against *Pectinophora gossypiella*

Formulations	LC_{50} (ppm)	Slope	Toxicity Index
1	1440.02	2.79	20.19
2	1009.7	1.811	28.79
3	1274.8	2.811	22.80
4	326.6	0.88	89.01
5	967.39	0.870	30.05
6	786.005	1.37	36.98
7	1561.93	2.79	18.61
8	290.73	0.66	100
9	967.39	0.870	30.05
10	1647.56	2.52	17.64

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