Pesticide formulation testing: importance and protocols

Thammali Hemadri, Hanchinal, S. G., Prabhuraj, A., Pavankumar K., Somashekhar Gaddanakeri, Shrikant, Meghana V Kodler, Archana, P., Sangeeta and Prakash A Jituri

Abstract

Analyses of pesticide formulations is critical in the backdrop of availability of quality pesticide market for ultimate benefit of farmers and ecosystem health. This article highlights the significance of comprehensive pesticide formulation analysis protocols, encompassing methods for determining active ingredient concentrations and assessing physical and chemical parameters. Employing advanced analytical techniques such as spectroscopy and chromatography for pesticide formulation analysis will eventually maintain the quality of pesticides for the sustainable management of pests.

Key words: Pesticide formulation, instrumental analysis, volumetric analysis, pest management

In India, over 330 pesticides are registered of which includes insecticides, fungicides, herbicides (DPPQS, 2023). The current market of non-genuine pesticide is INR 3,200 Cr (USD 525 Million) which constitutes 25 per cent by value and 30 per cent by volume of the total domestic market of agrochemicals in India as per Industry reports, primary interviews, news articles and Tata Strategic analysis (FICCI, 2015). Monitoring registration and regulation of pesticides is governed by the Pesticide Management Bill 2020, which replaced the Insecticide Act 1968 (DPPQS, 2023). The Insecticide Act of 1968 established the Central Insecticide Laboratory, Faridabad, under section 16 with two regional pesticide testing laboratories at Chandigarh and Kanpur (DPPQS, 2023). Current Quality Control of Pesticides in States/UTs during the last five years shows that 68078 pesticide samples were analysed (DPPQS, 2023).In the beginning, these laboratories involved in analysing pesticide samples drawn by any officer or the body authorized by the Central or State Governments and submit certificates of analysis to the concerned authority. Regional Pesticide Testing Laboratories (RPTLs) had a huge target of analysis of 1550 samples per annum that necessitated the establishment of 71 State Pesticide Testing Laboratories (SPTL) spread across India of which 14 are NABL accredited (DPPQS, 2023). As per the Pesticide Management Bill 2020, the routine process of pesticide formulations monitoring includes drawing of pesticide samples randomly from the market by and send it to SPTL for quality analysis. At SPTL, the insecticide analyst (Agriculture Officer/ Asst. Director of Agriculture) will perform the test and issue the certificate (Dileep Kumar and Narasimha Reddy, 2021). If the sample fails to meet the required standard, the manufacturer faces the consequences in a court of law (Dileep Kumar and Narasimha Reddy, 2021).

Pesticide formulation analysis main moto to find the substandard pesticide in the pesticide market. Substandard definition according to insecticide act, 1968 states that it does not conform to the active ingredient test approved for it by the Registration Committee and its active ingredient is within five percent of the nominal value when applied beyond the

S.NO.	PESTICIDE FORMULATION TESING	
1.	Instrumental	Volumetric
2.	HPLC	Precipitation Titration
3.	GC	Gravimetric Analysis
4.	MS	Emulsion Stability
5.	FTIR	Cold test
6.	NMR	Heat stability test
7.	UV-Vis. Spectroscopy	Persistent Foam
8.	Particle Size Analyser	Suspensibility test
9.	Thermal Analysis (DSC, TGA)	
10.	X-ray Diffraction (XRD)	
11.	Automatic Potentiometric Titrator	
12.	Karl Fischer Titrator	
13.	Sieve shaker	
14.	pH meter	
15.	Flash point apparatus	

Table 1: Types of pesticide formulation testing

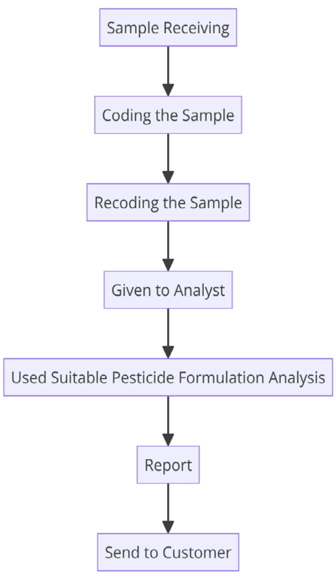

Fig. 1: Series of pesticide formulation analysis in the laboratory

upper and lower limits prescribed for conforming to the test, provided that no tolerance limit shall apply in case of pesticides, which are registered on minimum purity basis (FICCI, 2015).

Pesticide formulations can be analysed in two ways viz., Instrumental pesticide formulation analysis and volumetric pesticide formulation analysis (Table 1).

1. **Pesticide Formulation Analysis**

1.1 **Instrumental Pesticide Formulation Analysis**

Instrumental pesticide formulation analysis involves the use of advanced analytical techniques and instruments to evaluate the composition, properties, and quality of pesticide formulations. These techniques provide highly accurate and sensitive measurements, allowing researchers, regulators, and manufacturers to gain insights into the chemical and physical characteristics of pesticide products. Here, we'll delve into some of the key instrumental methods commonly used in pesticide formulation analysis:

1.1.1 **High-Performance Liquid Chromatography (HPLC):** HPLC is a versatile technique used to separate, identify, and quantify individual components within a pesticide formulation. It is especially effective for analysing active ingredients, impurities, and degradation products. HPLC employs a liquid mobile phase to transport the sample through a stationary phase, and detectors like UV provide information about the separated compounds.

Eg: Acetamiprid 20 % SP (IS 16328: 2017), Imidacloprid 17.8 % SL (IS 15443: 2004).

1.1.2 Gas Chromatography (GC): GC is employed for the analysis of volatile and thermally stable compounds. Pesticide formulations often include volatile solvents and certain active ingredients that can be effectively separated and quantified using this technique. GC utilizes a gaseous mobile phase to carry the sample through a stationary phase, and detectors like flame ionization detector (FID) or mass spectrometry are used to detect and quantify compounds.

Eg: Chlorpyrifos 20 % EC (IS 8944: 2005), Fenpropathrin 10 EC (IS 15161: 2002), Fenvalerate 11 % EC (IS 11997: 1987), Organochlorine, Organophosphorus, Pyrethroids *etc*. can be analyzed by GC

1.1.3 Mass Spectrometry (MS): Mass spectrometry is a powerful technique for identifying and quantifying compounds based on their mass-to-charge ratios. MS can be coupled with various instruments (such as GC-MS or LC-MS) to analyse metabolites from parent compounds, complex mixtures in pesticide formulations and identify unknown compounds.

Eg: Fipronil and its metabolite fipronil sulfone (Metabolite will be form after spraying of pesticide on plant leaf, soil, insect)

1.1.4 Fourier-Transform Infrared Spectroscopy (FTIR): FTIR spectroscopy measures the interaction of molecules with infrared light, providing information about functional groups present in the sample. It's useful for identifying different chemical groups in pesticide formulations and can help detect changes due to degradation or formulation changes.

Eg: Ethion (IS 10319: 1982)

The nominal value for pesticide active ingredient decided by the bureau of Indian standards as if percent pesticide formulation up to 9 % the range should be between + 10 to -5 and above 9 and below 50 the range should be + 5 to -5 and 50 and above it should be +5 and -3.

1.1.5 Nuclear Magnetic Resonance (NMR) Spectroscopy: NMR provides detailed information about molecular structure and composition by analysing the interactions of nuclei with a magnetic field. It's particularly useful for elucidating the structure of complex molecules, confirming the identity of active ingredients, and studying the interactions between different components in the formulation.

1.1.6 UV-Visible Spectroscopy: UV-Vis spectroscopy measures the absorption of ultraviolet and visible light by molecules. It's used to quantify the concentration of active ingredients and assess the stability of formulations by monitoring changes in absorption spectra over time.

Eg: Cartap hydrochloride (IS 14184: 1994), Monocrotophos, Glyphosate (IS 12502: 1988), Malathion EC (IS: 2567: 1978)

1.1.7 Particle Size Analyser (Dynamic Light Scattering, Laser Diffraction): Pesticide formulations often come in various forms, such as suspensions or emulsions, where particle size plays a critical role in stability and effectiveness. Techniques like dynamic light scattering and laser diffraction provide insights into particle size distribution, allowing manufacturers to optimize formulation properties.

Eg: DP/WP/Granular formulations

1.1.8 Thermal Analysis (DSC, TGA): Differential scanning calorimetry (DSC) and thermogravimetric analysis (TGA) provide insights into the thermal properties and stability of pesticide formulations. These techniques help identify melting points, decomposition temperatures, and potential thermal degradation pathways.

1.1.9 X-ray Diffraction (XRD): XRD is used to analyze the crystal structure of solid components in pesticide formulations. It's valuable for studying the arrangement of particles and assessing crystallinity changes that may occur during formulation or storage.

These instrumental methods offer a comprehensive understanding of pesticide formulations, including the identification, quantification, and characterization of active ingredients, inert components, and additives. The integration of these techniques ensures the quality, safety, and efficacy of pesticide products while aiding regulatory compliance and environmental protection efforts.

1.1.10 Automatic Potentiometric titrator: This instrument is used to find the acidity/alkalinity of pesticide formulation.

Eg: Chlorpyrifos – ≤ 0.05 percent by mass ((IS 8944: 2005)

1.1.11 Karl Fischer Titrator: This instrument is used to find the moisture of pesticide formulation.

Eg: Majority of technical formulations and some of formulations (Imidacloprid Technical moisture content should be ≤1 % (IS 15443: 2004)

1.1.12 Sieve shaker with test sieves of 2 mm, 1.7 mm, 1.4 mm

Sieve test: wet sieve test is a critical analytical technique in pesticide formulation because it ensures that the particle size distribution is controlled and consistent.

Eg: WP and Granular formulations

1.1.13 **pH meter: pH meter is an equipment to find the formulation pH**

Eg: Acetamiprid pH (Between 7.0-9.0) (IS 16328: 2017), Fipronil pH (Between 4.0-8.5) (IS 16145: ***2013)***

1.1.14 **Flash point apparatus (Abel)**

A flash point apparatus is a laboratory instrument used to determine the flash point of a flammable substance. The flash point is the lowest temperature at which the vapour of a substance can ignite in the presence of an open flame or spark. It is an important safety parameter for handling and storing flammable materials (IS 1448: 1960)

Here's how a flash point apparatus typically works:

Sample Preparation: A small quantity of the substance being tested (usually a few millilitres) is placed in a cup or container designed for the apparatus.

Test Procedure: The cup containing the sample is positioned in the apparatus, and it is typically exposed to an open flame or an electrical spark in a controlled environment.

Temperature Control: The temperature of the sample is gradually increased at a controlled rate (usually 1 or 2 degrees Celsius per minute) using a heating element.

Observation: An ignition source, such as a pilot flame, is directed towards the surface of the sample. The operator carefully observes the sample during the temperature ramp-up. The flash point is reached when a small flame or spark occurs at the surface of the sample, indicating that it has reached a temperature at which it can ignite.

Recording Data: The temperature at which the flash occurred is recorded as the flash point of the substance. The flash point of the material shall be above 24.5 C

Eg: EC and SL formulations

1.2 Volumetric pesticide formulation analysis

Volumetric analysis is a branch of analytical chemistry that involves measuring the volume of a solution of known concentration (titrant) required to react completely with a solution of the analyte, thus allowing for the determination of the analyte's concentration. In the context of pesticide formulation analysis, volumetric methods are often used to quantify the concentration of active ingredients or specific chemical components within the formulation. Volumetric analysis methods are widely utilized due to their simplicity, accuracy, and relatively low cost. Here, we'll elaborate on some of the key volumetric techniques used in pesticide formulation analysis:

1.2.1 Precipitation Titration: Precipitation titration involves the formation of an insoluble precipitate when a titrant is added to the analyte solution. This method can be used to determine ions that form insoluble compounds with specific reagents. In pesticide formulation analysis, precipitation titrations might be used to quantify certain ions or to detect impurities that form precipitates.

1.2.2 Gravimetric Analysis: While not strictly volumetric, gravimetric analysis is closely related. It involves the determination of the mass of a compound, which can then be used to calculate its concentration. Gravimetric methods are based on the principle that a specific compound can be separated and weighed to determine its concentration in the original sample. This technique can be used to analyse specific active ingredients or impurities in pesticide formulations.

1.2.3 Emulsion Stability: Emulsion stability is typically assessed using a 100 mL Crow receiver. The desired outcome is the observation of any phase separation, such as creaming at the top or sedimentation at the bottom, in a 100 mL emulsion created in standard hard water (342 ppm) with an EC volume not exceeding 2.0 mL (IS 6940: 1982).

Eg: EC and EW formulations (Chlorpyrifos 20 % EC, Fenpropathrin 10 % EC, Fenvalerate 11 % EC)

1.2.4 Cold test: Put 50mL of pesticide formulation in a 100mL transparent glass container and close it with a thermometer-equipped cork or stopper. Put the container in ice water to cool it to 10°C.When the pesticide in the container reaches 10°C, apply a little pesticide seeding crystal with minimal stopper opening in the shortest time. The formulation made from technical liquid pesticide does not need crystal seeding. Stir the material in the container gently at short intervals for one hour at 10°C. After one hour, check for turbidity or solid/oily matter separation (IS 6940: 1982).

Eg: All liquid formulations (Chlorpyrifos 20 % EC, Fenpropathrin 10 % EC, Fenvalerate 11 % EC, Imidacloprid 17.8 % SL)

1.2.5 Heat stability test: Place the 50 g of the sample in a bottle and if sample is volatile use sealed bottle. Seal the sample container hermetically and keep it in the oven at 55±2□C for 14 days. Remove the sample container from the oven, cool down to room temperature.

Eg: All liquid formulations

1.2.6 Persistent Foam: The sample mass is the mass needed to make 200 mL of a suspension with the concentration suggested in the product's instructions. When multiple concentrations are advised, choose the highest. Put 180 mL of standard hard water in the 250 mL measuring cylinder on a top pan balance and weigh in 1g of suspension concentrate. Fill to 200 mL with regular hard water. Stop the cylinder and invert 30 times. Start the stop watch after placing the stoppered cylinder upright on the bench. Read foam production and remaining after 1 min.

Eg: Fipronil 5 % SC (IS 16145: 2013)

1.2.7 Suspensibility test: The dry pesticide is slurred into 50 ml of test water in a 100 ml beaker. The slurry is quantitatively transferred to a 250 ml mixing cylinder is stoppered and inverted in 15 complete cycles. The mixing cylinder is allowed to stand for 30 minutes. After 30 minutes the top 225 ml is drawn off and the remaining suspension is dried. The residue weight will determine the percent suspensibility.

Eg: WP and SC formulations

Conclusion:

Pesticide formulation analysis (PFA) is a vital part of modern agriculture, assuring the efficacy, stability, and safety of the products. By employing advanced analytical techniques such as chromatography and spectroscopy, regulatory bodies can make decisions according to the act elimination of inferior quality pesticides and make use of quality pesticides to the farmers. It is important to note that though volumetric methods are valuable for many analyses, they might not be suitable for all types of pesticide formulations or analyses. For complex formulations with multiple active ingredients and additives, more sophisticated instrumental techniques like chromatography, spectroscopy, and mass spectrometry might be necessary to achieve accurate and comprehensive analysis. The substandard pesticides can be detected by PFA, which gives farmers the right to take compensation from the company according to consumer protection act, 2019. Therefore, the future implications of pesticide formulation analysis are significant and safeguarding the crop health. PFA ensures production of quality pesticides from industries, which in turn helps scientists to carry out quality research while evaluating pesticides against insect-pests before shortlisting them in university package of practices. It also creates trust among dealers and boosts their business.

References

Dileep Kumar A D, Narasimha Reddy, D. 2021. The Pesticide Management Bill 2020. Current science 121(3): 348-349. https://www.currentscience.ac.in/Volumes/121/03/0348.pdf

Directorate of Plant Protection, Quarantine & Storage (DPPQS); https://ppqs.gov.in/. (Accessed on 20.11.2023).

Federation of Indian Chambers of Commerce and Industry (FICCI), Study on Sub - Standard, Spurious/Counterfeit Pesticides in India 2015 - Report. Available from: https://croplife.org/wp-content/uploads/2015/10/Study-on-sub-standard-spurious-counterfeit-pesticides-in-India.pdf.

Indian Standard Methods of Test for Pesticides and Their Formulations, IS 6940: 1982, https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=6940&id=0. (Accessed on 20.11.2023).

Indian Standard of Acetamiprid, Soluble powder Specification, IS 16328: 2017, Available from: https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=16328&id=0.(Accessed on 20.11.2023).

Indian Standard of Chlorpyrifos, Emulsifiable concentrate Specification, IS 8944: 2005, Available from:https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=8944&id=0. (Accessed on 20.11.2023).

Indian Standard of Fenpropathrin Technical Specification, IS 15161: 2002, Available from: https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=IS+15161&id=0. (Accessed on 20.11.2023).

Indian Standard of Fenvalerate, Emulsifiable concentrate Specification, IS 11997: 1987, Availablefrom:https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=11997&id=0. (Accessed on 20.11.2023).

Indian Standard of Glyphosate Technical Specification, IS 12502: 1988, Available from: https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=11997&id=0. (Accessed on 18.11.2023).

Indian Standard of Imidacloprid, Technical Specification, IS 15443: 2004, Available from: https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=15443&id=0. (Accessed on 18.11.2023).

Indian Standard pesticide - Cartap Hydrochloride G specification, IS 14184: 1994, Available from:https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=14184&id=0. (Accessed on 15.11.2023).

Indian Standard specification for Ethion

Emulsifiable concentrates, IS 10319: 1982, Availablefrom: https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=IS+10319&id=0 (Accessed on 20.11.2023).

Specification for Malathion Emulsifiable concentrates, IS 2567: 1978, Available from: https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=2567&id=0. (Accessed on 18.11.2023).

Standard of Fipronil Suspension concentrate Specification, IS 16145: 2013, Available from https://standardsbis.bsbedge.com/BIS_SearchStandard.aspx?Standard_Number=16145&id=0. (Accessed on 11.11.2023).

AUTHORS

Thammali Hemadri*., Hanchinal, S. G., Prabhuraj, A., Pavankumar K., Somashekhar Gaddanakeri., Shrikant., Meghana V Kodler, Archana, P., Sangeeta

Pesticide Residue and Food Quality Analysis Laboratory, UAS, Raichur - 584104, Karnataka, India

Prakash A Jituri

Karnataka University, Dharwad – 580003, Karnataka, India

*Email: hemadriento123@gmail.com