

REVIEW ARTICLE

Hair Analysis for Pesticide Biomonitoring: Advances in Exposure Assessment and Forensic Applications

Anumita Mazumdar¹, Riya Reji², Aiswarya Jose³, Shristi Aich⁴

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ABSTRACT

Aim: Pesticide exposure poses significant public health risks due to environmental persistence, toxicity, and increasing cases of self-harm through pesticide ingestion. This review evaluates hair as a non-invasive matrix for pesticide biomonitoring, focusing on commonly detected pesticides, extraction and analytical techniques, and recent advancements for improved sensitivity and accuracy.

Methods: A systematic review was conducted on pesticide detection in hair, analyzing extraction and analytical techniques such as Gas Chromatography-Mass Spectrometry (GC-MS) and Liquid Chromatography-Mass Spectrometry (LC-MS). Methods were compared based on sensitivity, recovery, and precision to determine their suitability for exposure assessment and forensic investigations.

Conclusion: Hair serves as a valuable, non-invasive matrix for pesticide biomonitoring, aiding both environmental exposure studies and forensic toxicology. Continuous advancements in eco-friendly extraction methods and high-sensitivity detection techniques will further refine its applicability. Standardized protocols are essential for ensuring data comparability and enhancing the reliability of pesticide exposure assessments, particularly in cases of intentional poisoning.

KEYWORDS

• Pesticide • Biomonitoring • Sensitivity • Recovery • Precision • Agriculture
• Toxicology • Gas Chromatography-Mass Spectrometry • Liquid Chromatography-Mass Spectrometry

AUTHOR'S AFFILIATION:

¹ PG Student, Department of Forensic Science, Kristu Jayanti College (Autonomous), Bengaluru, Karnataka, India.

² PG Student, Department of Forensic Science, Kristu Jayanti College (Autonomous), Bengaluru, Karnataka, India.

³ PG Student, Department of Forensic Science, Kristu Jayanti College (Autonomous), Bengaluru, Karnataka, India.

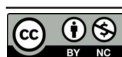
⁴ Assistant Professor, Department of Forensic Science, Jain University, Bengaluru, Karnataka, India.

CORRESPONDING AUTHOR:

Shristi Aich, Assistant Professor, Department of Forensic Science, Jain University, Bengaluru, Karnataka, India.

E-mail: shristiaich37807@gmail.com

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INTRODUCTION

Pesticides can potentially damage the ecosystem severely, particularly biodiversity. Pesticides are often applied in farming fields and seep into nearby water bodies, polluting rivers, lakes, and oceans. These runoffs can potentially disturb aquatic ecosystems, leading to such effects as algal blooms that reduce oxygen levels, killing fish and harming aquatic organisms.¹ In addition, the upper levels of trophism, e.g., birds and mammals that are supported by aquatic organisms for food, are threatened by the bioaccumulation of pesticides in the food chain.²

Based on the target species, pesticides are classified as insecticides, herbicides, fungicides, rodenticides, and fumigants. Insecticides such as carbamates and organophosphates inhibit acetylcholinesterase, leading to neurotoxicity.³ Paraquat is a herbicide that causes oxidative stress and pulmonary damage. Dithiocarbamates are some of the fungicides that can cause disruption of endocrine function.⁴ Rodenticides, including anticoagulants, interfere with blood coagulation and lead to bleeding. Methyl bromide and other fumigants have neurological effects. Long-term exposure to pesticides is linked to reproductive toxicity, neurological diseases, and carcinogenicity. To achieve risk assessment and regulatory measures to decrease health and environmental risks while sustaining agricultural effectiveness, it is essential to understand their toxicological profiles.⁵

Understanding the Risks of Long-Term Pesticide Exposure

Globally, pesticides are used extensively in public health to avert disease and in agriculture to protect crops. They have a broad spectrum of chemicals, mainly of the herbicide (e.g., acid herbicides), fungicide (e.g., azoles), insecticide (e.g., organochlorines (OCs), organophosphates (OPs), carbamates (CAs), pyrethroids (PYRs), and neonicotinoids (NEOs) categories.⁴

The Food and Agriculture Organisation of the United Nations Pesticides Use Report (2024) highlights a significant rise in global pesticide consumption, with agricultural use reaching 3.70 million tonnes in 2022, a 4% increase from 2021 and a 13% rise over the past decade. While Asia led pesticide

exports (3.5 Mt, valued at USD 21.7 billion), the Americas saw a 10% rise in usage, and Africa remained heavily reliant on imports. Conversely, Europe recorded a 5% decline since 1990, reflecting stricter regulations. Notably, biopesticides constituted 44% of total insecticide use in Oceania, underscoring a shift towards sustainable alternatives. These statistics emphasize the growing global dependency on pesticides, raising concerns about environmental and health impacts.⁶

Pesticide consumption in India has been a critical factor in agricultural sustainability, impacting crop yield, pest management, and environmental health. According to the Government of India's Ministry of Agriculture and Farmers Welfare, Directorate of Plant Protection, Quarantine and Storage (2024), there are significant regional disparities in the use of chemical and bio-pesticides across various states from 2018 to 2023. States like Maharashtra and Uttar Pradesh exhibit high dependency on chemical pesticides, whereas others show a gradual shift towards bio-pesticides. Analyzing these trends is essential for assessing the long-term effects of pesticide application on agriculture, ecological balance, and human health.⁷

Pesticide Poisoning as an Alarming Mode of Self-Harm

According to the National Crime Records Bureau (NCRB) Accidental Deaths and Suicides in India (ADSI) Report (2022), poisoning remains one of the leading methods of suicide in India, with a significant number of cases attributed to the consumption of insecticides. The data indicate that males account for a higher proportion of suicides by poisoning compared to females, reflecting potential socio-economic and psychological stressors. The overall trend suggests a gradual increase in such cases over the years, highlighting the urgent need for strict pesticide regulations, mental health interventions, and suicide prevention strategies. Addressing the easy availability of toxic substances and promoting awareness about their risks are crucial steps in mitigating this issue.⁸

Significance of Analysing Pesticides from Hair Samples

The most common fluids for biomonitoring exposure to contaminants are plasma and urine, which have been employed so far. These

fluids, however, only provide short-term data and can show a high level of variability in terms of pesticide levels, especially for nonpersistent chemicals. As a substitute for relying solely on these fluids, hair analysis has been proposed because it can provide information on chronic exposure averaged over several months. Short detection time windows and drastic variation in urinary chemical concentration result from the rapid excretion of most compounds out of urine between successive episodes of exposure. New approaches involving alternative matrices, such as hair, have been suggested to bypass the limitations of conventional biological matrices.⁸

Mechanisms of Pesticide Deposition in Hair

The entire length of the hair extends from the bulb rooted in the follicle through the shaft and ends at the tip. Three layers comprise the shaft: the medulla, cortex, and cuticle. Besides being somewhat resistant to chemical breakdown, cuticle can preserve its structural features for a more extended period. Beneath the cuticle's protective layer lies the second layer, known as the cortex. This part of the cortex mainly holds importance in the forensic field in relation to embedding pigment granules responsible for its distinct coloration.¹⁰

By blood flow, sebum, and environmental deposition, pesticides and metabolites reach hair. Hair sample is less labor-intensive, painless, and non-invasive compared to tissue biopsies or blood sampling. Since it is less affected by changes in metabolism, degradation, and environmental factors, it is more consistent than blood or urine.¹¹

Distinguishing Chronic vs. Acute Exposure

It serves as a valuable forensic tool in determining whether pesticide poisoning was a result of long-term exposure or acute ingestion, helping to establish intent in suicide cases. The presence of pesticides along the entire hair length suggests chronic exposure, indicating occupational or environmental contact over time. In contrast, if pesticides are detected only near the root, it points to recent ingestion, supporting the case of deliberate self-poisoning.^{10,11}

Determining the Poisoning Timeline

Since hair grows at an average rate of ~1 cm per month, forensic scientists can analyze the exposure history over time. This timeline helps

distinguish between chronic poisoning (due to repeated exposure) and a single fatal dose (indicative of suicide or homicide).^{10,11}

In forensic and toxicological studies, washing steps are often included to differentiate external contamination from ingested or systemic exposure, ensuring that detected pesticides are truly incorporated into the hair rather than just surface contaminants. The decontamination procedure depends on the specific analyte and should not alter the hair matrix. In the study "Hair decontamination procedure prior to multi-class pesticide analysis," researchers developed a washing protocol to effectively remove external pesticide contamination from hair samples. The procedure involves sequential washing with sodium dodecyl sulfate (SDS) and methanol, which was found to efficiently eliminate surface residues without affecting pesticides incorporated within the hair shaft. This approach ensures that subsequent analyses reflect systemic exposure rather than external contamination.¹²

Techniques for Pesticide Extraction From Hair Samples

Several innovative techniques have emerged as alternatives to traditional pesticide extraction methods, offering improved efficiency, sensitivity, and environmental sustainability. While several extraction techniques have been employed for hair analysis, they are predominantly used in cosmetic and pharmaceutical studies rather than pesticide residue analysis. Emerging green technologies like Deep Eutectic Solvents (DES) have shown promise in drug extraction, highlighting a vast potential for future studies in pesticide detection from hair samples. These include Matrix Solid Phase Dispersion (MSPD), Dispersive Liquid-Liquid Microextraction (DLLME), Ultrasound-Assisted Extraction (UAE), Supercritical Fluid Extraction (SFE), Microwave-Assisted Extraction (MAE), and Deep Eutectic Solvents (DES). The advantages and disadvantages have been mentioned in the Table. However, they have not been utilised for the extraction of hair samples. The most widely used analytical techniques for pesticide extraction include Liquid-Liquid Extraction (LLE), Solid-Phase Microextraction (SPME), and QuEChERS. Each method offers distinct advantages and limitations, making them

suitable for different applications.¹³

Liquid-Liquid Extraction (LLE)

A separation technique where analytes are transferred between two immiscible liquid phases, typically an aqueous phase and an organic solvent. Vigorous mixing allows analytes to migrate into the organic phase, which is then collected for analysis. While simple and versatile, LLE is time-consuming and requires large solvent volumes.¹⁴

Solid-Phase Microextraction (SPME)

Utilizes a fused silica or metal fiber coated with a stationary phase to extract analytes from gaseous, liquid, or headspace samples. SPME combined with GC-MS/MS is a highly sensitive method for analyzing multi-class pesticides in human hair.¹³ Recent advancements involve nanostructured sorbents such as graphene oxide, carbon nanotubes, and metal oxide nanoparticle, improving extraction efficiency. Additionally, magnetic solid-phase extraction (MSPE) eliminates the need for centrifugation and filtration by allowing phase separation using an external magnetic field. However, SPME fibers are costly, degrade with use, and show variability in analyte enrichment due to differences in coating thickness.¹⁴

Quenchers vs. QuEChERS: Quenchers refer to molecules or nanomaterials that absorb and dissipate fluorescence signals, often used in biosensors and molecular assays. In contrast, QuEChERS (Quick, Easy, Cheap, Effective, Rugged, and Safe) is a widely used pesticide extraction technique designed for food and environmental samples. The QuEChERS method replaces traditional complex extraction techniques by using acetonitrile instead of acetone, improving phase separation and efficiency after salt addition. It effectively removes polar pigments, fatty acids, organic compounds, and sugars, enhancing analytical accuracy.¹⁷ Recent advancements in QuEChERS techniques include Nano-Quenchers, where Gold Nanoparticle (AuNPs) and Graphene Oxide (GO) improve biosensor sensitivity due to their large surface area and high energy transfer efficiency, enabling label-free detection of DNA, RNA, and pesticide residues.¹⁶ Molecular quenchers such as Black Hole Quenchers (BHQs) and Iowa Black Quenchers (IBQs) absorb fluorescence without generating background signals, making them ideal for qPCR-based genetic analysis.¹⁵ Additionally,

Multiplex Quenching has enabled high-throughput detection, allowing multi-color quenchers to interact with various fluorophores for simultaneous target identification, with applications in disease diagnosis, food safety testing, and environmental monitoring¹⁸

Analytical Techniques

The most commonly used techniques for pesticide detection in hair, based on our review, include GC-MS, LC-MS/MS, HRMS, SERS, SPME-GC-MS, and FTIR, as listed in Table 1. These methods have been widely employed due to their sensitivity, specificity, and ability to detect both volatile and non-volatile pesticides. However, method development may vary depending on the pesticide class and the physicochemical properties of individual pesticides, requiring different extraction and detection strategies. The validation of these methods is typically conducted based on seven key factors: precision, accuracy, sensitivity, specificity, limit of detection (LOD), limit of quantification (LOQ), and reproducibility.^{13,17}

Summary of Analytical Techniques for Hair-Based Pesticide Detection across Seven Studies

In our review, we have compared seven studies that have employed these techniques, evaluating their analytical performance based on LOQ, precision, and accuracy. This comparative analysis provides insights into the effectiveness of different methods and highlights the need for further optimization and standardization in pesticide residue analysis from hair samples. MS/MS and GC-MS, with solid-phase microextraction (SPME) and QuEChERS being the most common extraction methods. Human hair was the primary sample type, with only one study analyzing bat hair, highlighting the limited research on pesticide exposure in animals despite their susceptibility to adverse health effects. LOD and LOQ values varied significantly, with lower limits observed in studies using UPLC-MS/MS, demonstrating its superior sensitivity. The Box chart in Figure 3 explains the distribution of key analytical parameters for pesticide detection in hair. Limit of Detection LOD, LOQ show low values but have high outliers (~250 and ~500 pg/mg), indicating sensitivity issues in some methods. Recovery is high (~120%) with minimal variability, while Precision (%RSD) ranges from 14-18%, with some variability (~22%). Overall, the data suggests strong recovery and

precision but highlights sensitivity challenges, emphasizing the need for optimized detection techniques.^{13,35-39}

The increasing application of advanced microextraction methods and high-resolution mass spectrometry demonstrates growing efforts to enhance sensitivity, reduce sample preparation complexity, and improve pesticide biomonitoring in forensic and environmental toxicology.

DISCUSSION

Hair analysis can help fill gaps in the monitoring frameworks that exist today. Research on hair analysis for pesticide monitoring has gained momentum. It makes retrospective exposure assessment possible, which is a critical component of researching long-term illnesses, including cancer, neurological diseases, and developmental disabilities that are often linked to pesticide exposure. In addition, hair analysis may provide insight into differences in exposure among different populations, especially vulnerable groups such as children and farmworkers. Although these procedures offer advantages, issues regarding the standardization of analytical procedures, data interpretation, and distinguishing between endogenous pesticide residues and external contamination exist. To establish hair analysis as a reliable tool, these constraints must be resolved with advanced analytical techniques, such as LC-MS, and standardised methods need to be developed.¹⁹

Most analytical and extraction techniques have been utilized for hair samples; however, their application to pesticide extraction remains limited. Despite advancements in nanotechnology for pesticide analysis, research on the application of novel nanoparticle in hair analysis remains limited. Few studies have comprehensively reported key analytical parameters such as sensitivity, detection limits (LOD/LOQ), and accuracy, raising concerns about reliability. Ethical considerations surrounding the collection of human and animal hair samples further restrict large-scale studies. While researchers are increasingly exploring greener, less invasive techniques, challenges in standardization and external contamination persist. The future scope remains promising, with the need for more robust investigations into nano-based analytical enhancements to improve precision and applicability in pesticide exposure assessment.²⁰

CONCLUSION

Exposure to pesticides remains a significant concern, particularly for farmers and agricultural workers who face chronic exposure through inhalation, dermal absorption, and ingestion. Additionally, the rising trend of pesticide ingestion for self-harm has become a public health issue, necessitating improved monitoring methods. Hair, being a non-invasive and long-term biomarker, remains a promising tool for exposure assessment. Beyond routine monitoring, hair analysis can also aid forensic investigations, as pesticide residues in hair persist even after death, helping determine the cause of death in cases of poisoning while also slowing decomposition, preserving critical evidence. Despite its potential, further research is needed to enhance analytical precision using advanced technologies, and further expanding research on animal hair analysis could provide valuable insights into environmental pesticide exposure and its impact on ecosystems.

Challenges of Hair Analysis for Pesticide Exposure

Biological Variability

Hair analysis reflects long-term rather than short-term exposure. Individual differences in hair growth, influenced by factors like genetics and health, make it difficult to establish precise exposure timelines. Pesticide absorption varies by chemical properties and exposure route, affecting consistency.⁹

Sample Contamination

Hair can absorb external contaminants from the environment or surfaces, requiring careful cleaning before analysis. While washing reduces contamination, complete removal can be challenging.²¹

Standardization Issues

Detection sensitivity varies among pesticides, with some being harder to measure due to low incorporation or chemical instability. Hair analysis provides exposure insights but does not always quantify exact levels.²¹

Ethical Concerns

Hair collection raises privacy considerations, especially regarding consent. Cultural factors may also influence acceptance in some regions.²²

Conflict of Interest

The authors declare that there is no conflict of interest.

Table 1: Comparison of various extraction techniques

Technique	Description	Advantages	Disadvantages	Citations
QuEChERS (Quick, Easy, Cheap, Effective, Rugged, and Safe)	A solvent-based extraction method for multi-residue pesticide analysis	High recovery, reduced solvent usage, minimal sample handling	May require further purification steps	24
Solid Phase Microextraction (SPME)	Uses a coated fiber to extract pesticides directly from the sample	Solvent-free, rapid, highly sensitive	Fiber lifetime is short and needs replacement	14
Matrix Solid Phase Dispersion (MSPD)	Combines sample homogenization, extraction, and cleanup in one step	Reduces solvent use and processing time	Requires optimization for different matrices	14
Dispersive Liquid-Liquid Microextraction (DLLME)	Uses a small volume of extractant to isolate pesticides from aqueous samples	High enrichment factor, rapid, low detection limits	Limited to hydrophobic analytes	13
Ultrasound-Assisted Extraction (UAE)	Uses ultrasonic waves to enhance pesticide extraction efficiency	Faster extraction, improved recovery, reduced solvent use	Requires careful optimization of parameters	26
Supercritical Fluid Extraction (SFE)	Uses supercritical CO ₂ to extract pesticides from samples	Non-toxic, highly selective for non-polar pesticides	Expensive instrumentation	28
Microwave-Assisted Extraction (MAE)	Uses microwave energy to improve pesticide extraction efficiency	Faster, improved recovery rates	Requires specialized equipment	29
Deep Eutectic Solvents (DES)	Uses green solvents for pesticide extraction with high efficiency	Environmentally friendly, tunable properties	Requires further validation for widespread application	27

Table 2: Different analytical techniques used for hair pesticide long-term monitoring, along with their advantages and disadvantages

Technique	Description	Qualitative/ Quantitative	Advantages	Disadvantages	Citations
Gas Chromatography-Mass Spectrometry (GC-MS)	Separates and identifies pesticide residues in hair based on mass spectra, commonly used for volatile pesticides	Quantitative	High sensitivity, reliable identification of organophosphates and pyrethroids	Requires extensive sample preparation, expensive instrumentation	13
Liquid Chromatography-Mass Spectrometry (LC-MS/MS)	Detects non-volatile and polar pesticides in hair with high accuracy	Quantitative	High specificity, suitable for multi-residue pesticide analysis	Requires careful sample preparation and matrix effect control	9,13
High-Resolution Mass Spectrometry (HRMS)	Provides precise mass identification of pesticide residues in hair, used for retrospective exposure assessment	Quantitative	Extremely accurate and specific for long-term pesticide monitoring	Expensive instrumentation and requires high technical expertise	30
Surface-Enhanced Raman Spectroscopy (SERS)	Uses nanopArticle to enhance Raman signals for pesticide detection in hair. *Technique has no direct link for hair samples but can be explored for the same.	Qualitative	Minimal sample preparation, rapid and non-destructive	Requires optimization for different pesticide classes	31
Solid Phase Microextraction (SPME) coupled with GC-MS	Uses a coated fiber to extract pesticides from hair before GC-MS analysis	Quantitative	Solvent-free, enhances extraction efficiency, suitable for volatile pesticides	Limited fiber lifespan and variable extraction efficiency	32
Fourier Transform Infrared Spectroscopy (FTIR)	Identifies functional groups of pesticide residues in hair based on infrared absorption patterns	Qualitative	Non-destructive, rapid analysis, requires minimal sample	Requires a reference spectral library for accurate interpretation	33
UPLC-MS/MS (Ultra-Performance Liquid Chromatography - Tandem Mass Spectrometry)	Used for high-resolution and trace-level pesticide detection in hair. Often used in biomonitoring studies.	Quantitative	High sensitivity and fast analysis, Suitable for detecting multiple pesticides at low concentrations	Expensive instrumentation and Requires expertise in sample preparation	34

Table 3: Key studies on pesticide biomonitoring using hair as a matrix.

All LOD/LOQ values were reported in ng/g hair (equivalent to pg/mg), the standard unit for solid biological matrices like hair. No conversions from solution-based concentrations (e.g., ng/mL or µg/L) were included unless they could be directly traced to hair mass.

Study	Analytical Method	Extraction Method	Sample Type	Pesticides Analyzed	LOD (ng/g)	LOQ (ng/g)	% Recovery	Precision (%RSD)	Publication Date
Analysis of pesticides in human hair by SPME-GC-MS/ ¹³	GC-MS	Solid Phase Microextraction	Human Hair	Acetochlor, Alachlor, Benfuracarb, Bifenthrin, Dicofof	0.5-1	2-2	NA	NA	2011
Development and application of GC-MS method for monitoring of long-term exposure to the pesticide cypermethrin. ³⁸	GC-MS	LLE + Derivatization	Human Hair	Pyrethroid Metabolites (cis/trans-Cl ₂ CA, 3-PBA)	1.0-4.0	3.3-13.3	84.8-96.4% ± 7.8-20.4	4.1-9.7%	2014
Assessment of human exposure to pesticides by hair analysis: The case of vegetable-producing areas in Burkina Faso. ³⁵	GC-MS, UPLC-MS/MS	d-SPE QuEChERS	Human Hair	Acetochlor, Chlordane, Chlorpyrifos, λ-Cyhalothrin	0.6-20.6	2-68.5	52.4-118.7%	6.9-22.4%	2017
Development of an LC-MS/MS method for non-invasive biomonitoring of neonicotinoid and systemic herbicide pesticide residues in bat hair. ³⁶	LC-MS/MS	Solid Phase Microextraction	Bat Hair	2,4-D, Atrazine, Dicamba, Glyphosate, Carbaryl, Clothianidin, Imidacloprid, Thiamethoxam	0.12-27.7	0.36-84.0	70.8-121.0%	1.1-13.2%	2022
Rapid LC-MS/MS quantification of Organophosphate non-specific metabolites in hair using alkaline extraction approach. ³⁹	LC-MS/MS	Alkaline Methanol Extraction	Human Hair	OP Metabolites (DMP, DEP, DETP)	0.5-2.0	1.5-5.0	65-119%	2.6-10.4%	2023
Exposure to pesticides, persistent and non-persistent pollutants in French 3.5-year-old children: Findings from comprehensive hair analysis in the ELFE national birth cohort. ³⁷	LC-MS/MS, GC-MS/MS	Dual-phase solvent	Human Hair (3.5 y/o children, n=222)	Organophosphates, Pyrethroids, Carbamates, Herbicides	1-25	5-70	75-120%	3-15%	2024

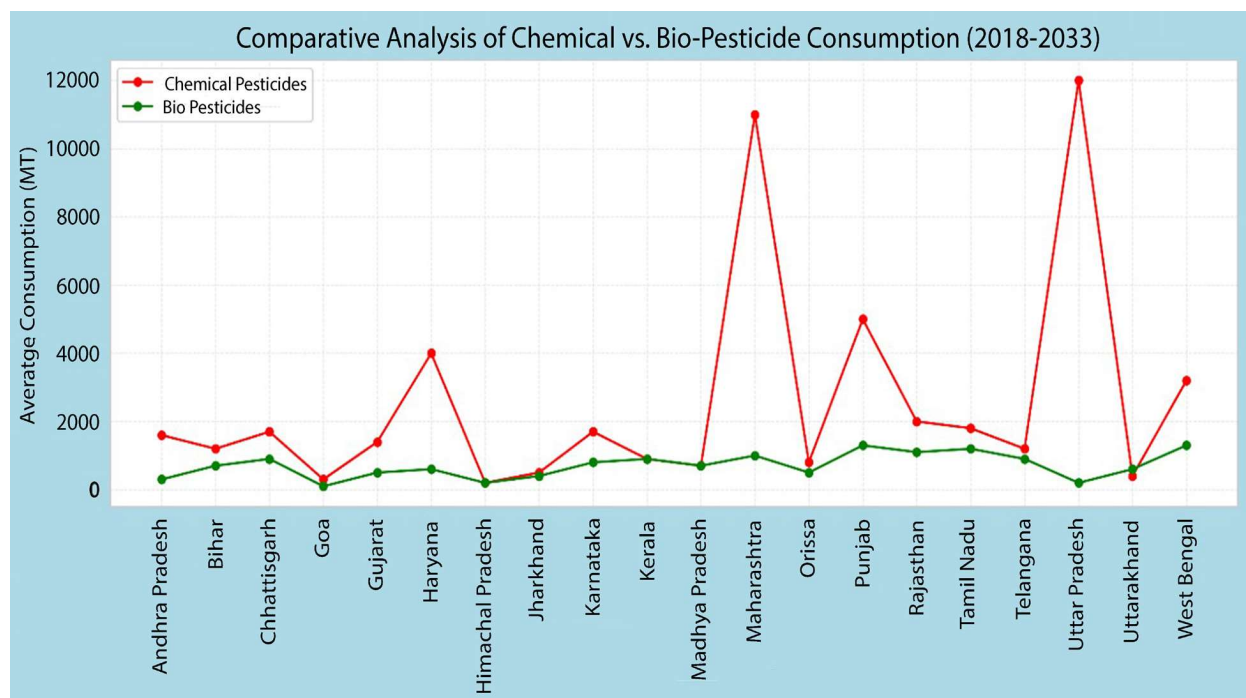


Figure 1: The figure presents the average quantity (in metric tons) of chemical and bio-pesticides used across different Indian states between 2018 and 2023. The red line represents chemical pesticide usage, while the green line represents bio-pesticide usage (*Source:* Ministry of Agriculture and Farmers Welfare, Government of India. *Pesticide consumption data:* PPQS, 2024. Available from: <https://ppqs.gov.in>)

DISTRIBUTION OF SUICIDES BY CONSUMING INSECTICIDES

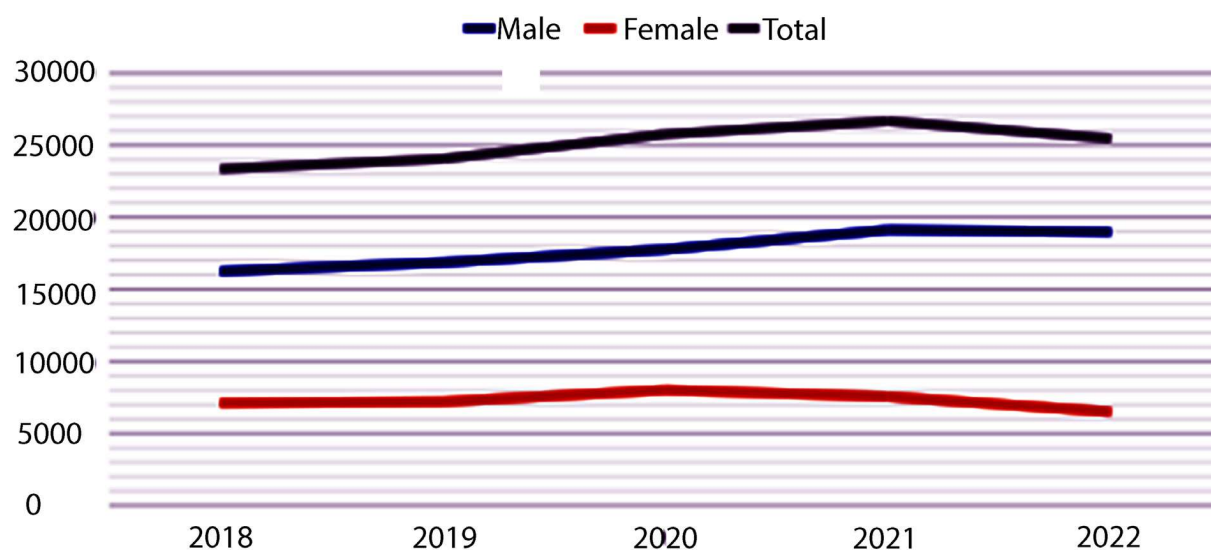


Figure 2: The graph illustrates the trend in suicides due to insecticide consumption from 2018 to 2022, categorized by gender (*Source:* National Crime Records Bureau. *ADSI Report 2022*. Available from: <https://ncrb.gov.in/en/accidental-deaths-suicides-india>)

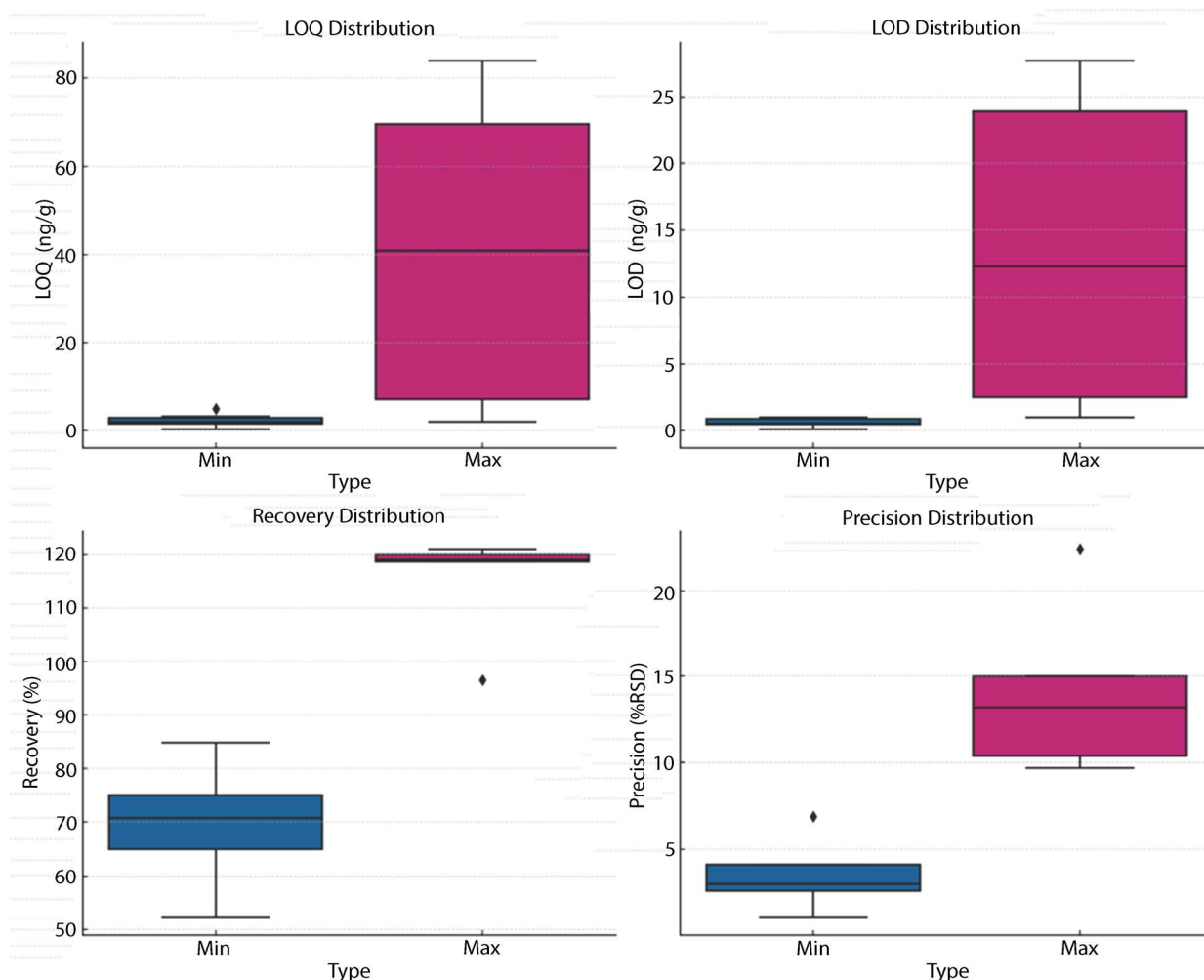


Figure 3: Boxplot Representation of LOQ, LOD, Recovery, and Precision Distributions

All LOD/LOQ values were reported in ng/g hair (equivalent to pg/mg), the standard unit for solid biological matrices like hair. No conversions from solution-based concentrations (e.g., ng/mL or $\mu\text{g/L}$) were included unless they could be directly traced to hair mass.

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Abbreviations:

1. **GC-MS** - Gas Chromatography-Mass Spectrometry
2. **LC-MS** - Liquid Chromatography-Mass Spectrometry
3. **LC-MS/MS** - Liquid Chromatography-Tandem Mass Spectrometry
4. **HRMS** - High-Resolution Mass Spectrometry
5. **SERS** - Surface-Enhanced Raman Spectroscopy
6. **SPME** - Solid-Phase Microextraction
7. **FTIR** - Fourier Transform Infrared Spectroscopy
8. **UPLC-MS/MS** - Ultra-Performance Liquid Chromatography-Tandem Mass Spectrometry
9. **LLE** - Liquid-Liquid Extraction
10. **DLLME** - Dispersive Liquid-Liquid Microextraction
11. **UAE** - Ultrasound-Assisted Extraction
12. **SFE** - Supercritical Fluid Extraction
13. **MAE** - Microwave-Assisted Extraction
14. **DES** - Deep Eutectic Solvents
15. **MSPD** - Matrix Solid Phase Dispersion
16. **QuEChERS** - Quick, Easy, Cheap, Effective, Rugged, and Safe
17. **LOD** - Limit of Detection
18. **LOQ** - Limit of Quantification
19. **%RSD** - Percentage Relative Standard Deviation
20. **OPs** - Organophosphates
21. **OCs** - Organochlorines
22. **CAs** - Carbamates

23. **PYRs** - Pyrethroids
24. **NEOs** - Neonicotinoids
25. **SDS** - Sodium Dodecyl Sulfate
26. **AuNPs** - Gold NanopArticle
27. **GO** - Graphene Oxide
28. **BHQs** - Black Hole Quenchers
29. **IBQs** - Iowa Black Quenchers
30. **qPCR** - Quantitative Polymerase Chain Reaction

REFERENCES

1. Paerl H.W. Mitigating toxic planktonic cyanobacterial blooms in aquatic ecosystems. *Et al* 2018; 10(2): 76. <https://doi.org/10.3390/toxins10020076>
2. McMahon T.A., Halstead N.T., Johnson S., *et al.* Fungicide-induced declines of freshwater biodiversity modify ecosystem functions. *Et al* 2012; 15(7): 714–22. <https://doi.org/10.1111/j.1461-0248.2012.01790.x>
3. Casida J.E. Pest toxicology: The primary mechanisms of pesticide action. *Et al* 2009; 22(4): 609–19. <https://doi.org/10.1021/tx8004949>
4. Sharma B., Agrawal A. Pesticides induced oxidative stress in mammals: a review. *Et al* 2010; 1(3): 90–104. <https://www.researchgate.net/publication/202037053>
5. Gupta R.C., editor. *Et al.* Elsevier; 2011.
6. <https://www.sciencedirect.com/book/9780128001592>
7. Food and Agriculture Organization of the United Nations. FAOSTAT: Pesticides use dataset. Rome: FAO; 2022 cited 2025 Jul 1. Available from: <https://www.fao.org/faostat/en/#data/RP>
8. Ministry of Agriculture and Farmers Welfare, Government of India. Pesticide consumption data: Directorate of Plant Protection, Quarantine & Storage (PPQS). Faridabad: PPQS; 2024 cited 2025 Jul 1. Available from: <https://ppqs.gov.in>
9. National Crime Records Bureau, Ministry of Home Affairs, Government of India. *Et al* Internet. New Delhi: NCRB; 2023 cited 2025 Jul 1. Available from: <https://ncrb.gov.in/en/accidental-deaths-suicides-india>
10. Appenzeller B.M.R., Hardy E.M., Grova N., Chata C., Fayš F., Briand O., *et al.* Hair analysis for the biomonitoring of pesticide exposure: comparison with blood and urine in a rat model. *Archives of Toxicology* Internet. 2016 Dec 23; 91(8): 2813–25. Available from: <https://doi.org/10.1007/s00204-016-1910-9>
11. Airlie M., Robertson J., Brooks E. Forensic hair analysis – Worldwide survey results. *Forensic Science International* Internet. 2021 Aug 20; 327: 110966. Available from: <https://doi.org/10.1016/j.forsciint.2021.110966>
12. Tsatsakis A., Tzatzarakis M., Tutudaki M., Babatsikou F., Alegakis A., Koutis C. Assessment of levels of organochlorine pesticides and their metabolites in the hair of a Greek rural human population. *Human & Experimental Toxicology* Internet. 2008 Dec 1; 27(12): 933–40. Available from: <https://doi.org/10.1177/0960327108102047>
13. Duca R., Hardy E., Salqu bre G., Appenzeller BMR. Hair decontamination procedure prior to multi-class pesticide analysis. *Drug Testing and Analysis* Internet. 2014 May 9; 6(S1): 55–66. Available from: <https://doi.org/10.1002/dta.1649>
14. Schummer C., Appenzeller B.M.R., Millet M., Wennig R. Analysis of pesticides in human hair by SPME-GC-MS/MS. *Et al* 2011; 1218(43): 7615–21. <https://doi.org/10.1016/j.chroma.2011.07.062>
15. Duca R.C., Salquebre G., Hardy E., Appenzeller BMR. Comparison of solid phase- and liquid/ liquid-extraction for the purification of hair extract prior to multi-class pesticides analysis. *Journal of Chromatography B* Internet. 2014 Mar 1; 955–956: 98–107. Available from: <https://doi.org/10.1016/j.jchromb.2014.02.035>
16. Kwon N.H., Lee Y.R., Kim H.S., Cheong J.C., Kim J.Y. Hybrid Solid-Phase extraction for selective determination of methamphetamine and amphetamine in dyed hair by using gas Chromatography–Mass spectrometry. *Molecules* Internet. 2019 Jul 9; 24(13): 2501. Available from: <https://doi.org/10.3390/molecules24132501>
17. Karnwal A., Sachan RSK, Devgon I., Devgon J., Pant G., Panchpuri M., *et al.* Gold nanopArticle in nanobiotechnology: From synthesis to biosensing applications. *ACS Omega* Internet. 2024 Jul 5; 9(28): 29966–82. Available from: <https://doi.org/10.1021/acsomega.3c10352>
18. Sidstedt, M., R dstr m, P., and Hedman, J. PCR inhibition in qPCR, dPCR and MPS-mechanisms and solutions. *Et al*, 2020; *et al.* (9): 2009–2023. <https://doi.org/10.1007/s00216-020-02490-2>
19. Li Z., Dong H., Wang H., Li D., Zhou S., Zhai S., *et al.* Novel time-resolved fluorescent-based multiplex immunochromatography test strip

- for simultaneous detection of pesticides in vegetables. *Food Chemistry Internet*. 2024 Nov 5; 464: 141916. Available from: <https://doi.org/10.1016/j.foodchem.2024.141916>
20. Taira K., Aebi A., Glauser G., Mitchell EAD. Urinary neonicotinoids in rural population of the Philippines and comparison with paired hair samples. *Et al.* 2023 cited 2025 Jul 1. Available from: <https://www.researchgate.net/publication/380698769>
21. Zareef I., Riaz A., Salahuddin, Fatima M., Anwar A., Khan S., et al. Unveiling the power of nanomaterials in the area of forensics. In: *Et al Internet*. Cham: Springer; 2024 cited 2025 Jul 1. p. 25–54. Available from: https://link.springer.com/chapter/10.1007/978-3-031-57843-4_2
22. Morton J., Carolan V.A., Gardiner PHE. Removal of exogenously bound elements from human hair by various washing procedures and determination by inductively coupled plasma mass spectrometry. *Analytica Chimica Acta Internet*. 2002 Mar 1; 455(1): 23–34. Available from: [https://doi.org/10.1016/s0003-2670\(01\)01578-1](https://doi.org/10.1016/s0003-2670(01)01578-1)
23. Thompson N., McGill T., Bunn A., Alexander R. Cultural factors and the role of privacy concerns in acceptance of government surveillance. *Journal of the Association for Information Science and Technology Internet*. 2020 Jun 25; 71(9): 1129–42. Available from: <https://doi.org/10.1002/asi.24372>
24. Wei, J., Cao, J., Tian, K., Hu, Y., Su, H., Wan, J., and Li, P. Trace determination of five organophosphorus pesticides by using QuEChERS coupled with dispersive liquid-liquid microextraction and stacking before micellar electrokinetic chromatography. *Analytical Methods*, 2015; 7(14): 5801–5807. <https://doi.org/10.1039/c5ay00692a>
25. Kim, J., Cho, H., Suh, J. H., Lee, J., Lee, E., Jin, C. H., et al. Analysis of nicotine metabolites in hair and nails using QUECHERS method followed by liquid Chromatography-Tandem Mass spectrometry. *Et al*, 2020; et al. (8): 1763. <https://doi.org/10.3390/molecules25081763>
26. Sharafi, K., Fattahi, N., Pirsahab, M., Yarmohamadi, H., and Davil, M. F. Trace determination of lead in lipsticks and hair dyes using microwave-assisted dispersive liquid-liquid microextraction and graphite furnace atomic absorption spectrometry. *Et al*, 2015; et al. (5): 489–95. <https://doi.org/10.1111/ics.12221>
27. Zhong, Z., Li, G., Wu, R., Zhu, B., and Luo, Z. Determination of aminophenols and phenol in hair colorants by ultrasound-assisted solid-phase dispersion extraction coupled with ion chromatography. *Et al*, 2014; et al. (16): 2208–2214. <https://doi.org/10.1002/jssc.201301252>
28. Wang, D., Yang, X., Tang, R., and Yao, F. Extraction of Keratin from Rabbit Hair by a Deep Eutectic Solvent and its Characterization. *Et al*, 2018; et al. (9): 993. <https://doi.org/10.3390/polym10090993>
29. Lehotay S.J. Supercritical fluid extraction of pesticides in foods. *Journal of Chromatography A Internet*. 1997 Oct 1; 785(1–2): 289–312. Available from: [https://doi.org/10.1016/s0021-9673\(97\)00461-5](https://doi.org/10.1016/s0021-9673(97)00461-5)
30. Llompart M., Celeiro M., Garcia-Jares C, Dagnac T. Microwave-Assisted extraction of pesticides and emerging pollutants in the environment. In: *Comprehensive analytical chemistry Internet*. 2017. p. 131–201. Available from: <https://doi.org/10.1016/bs.coac.2017.01.004>
31. Gkountouras D., Boti V., Albanis T. High resolution mass spectrometry targeted analysis and suspect screening of pesticide residues in fruit samples and assessment of dietary exposure. *Environmental Pollution Internet*. 2024 May 10; 352: 124143. Available from: <https://doi.org/10.1016/j.envpol.2024.124143>
32. Bakar NA, Fronzi M, Shapter JG. Surface-Enhanced RAMAN spectroscopy using a silver nanostar substrate for neonicotinoid pesticides detection. *Sensors Internet*. 2024 Jan 8; 24(2): 373. Available from: <https://doi.org/10.3390/s24020373>
33. Aspromonte J., Lancioni C., Purcaro G. Solid-Phase Microextraction—Gas Chromatography Analytical Strategies for pesticide analysis. *Methods and Protocols Internet*. 2022 Oct 17; 5(5): 82. Available from: <https://doi.org/10.3390/mps5050082>
34. Mohamed B.A., Janaki P. Determination of active ingredients in commercial insecticides using spectral characteristics of Fourier transform infrared spectroscopy (FTIR). *Journal of Applied and Natural Science Internet*. 2021 Jul 19; 13(SI): 110–23. Available from: <https://doi.org/10.31018/jans.v13isi.2809>
35. Kovalczuk T., Jech M., Poustka J., Hajšlová J. Ultra-performance liquid chromatography-tandem mass spectrometry: A novel challenge in multiresidue pesticide analysis in food. *Analytica Chimica Acta Internet*. 2006 Jun

- 19; 577(1): 8-17. Available from: <https://doi.org/10.1016/j.aca.2006.06.023>
36. Lehmann, E., Oltramare, C., Dibié, J.N., Konaté, Y., and De Alencastro, L.F. Assessment of human exposure to pesticides by hair analysis: The case of vegetable-producing areas in Burkina Faso. *Environment International*, 2017; 111: 317-31. <https://doi.org/10.1016/j.envint.2017.10.025>
37. Hooper S.E., Amelon S.K., Lin C.H. Development of an LC-MS/MS method for non-invasive biomonitoring of neonicotinoid and systemic herbicide pesticide residues in bat hair. *Et al.* 2022; 10(2): 73.
38. Available from: <https://www.mdpi.com/2305-6304/10/2/73>
39. Macheka L.R., Palazzi P., Iglesias-González A, *et al.* Exposure to pesticides, persistent and non-persistent pollutants in French 3.5-year-old children: Findings from comprehensive hair analysis in the ELFE study. *Et al.* 2024; 190: 107848. Available from: <https://www.sciencedirect.com/science/article/pii/S0160412024004677>
40. Kavvalakis M.P., Tzatzarakis M.N., Alegakis A.K., *et al.* Development and application of a GC-MS method for monitoring long-term exposure to the pesticide cypermethrin in hair samples. *Et al.* 2014; 6(5): 440-446. Available from: <https://analyticalsciencejournals.onlinelibrary.wiley.com/doi/abs/10.1002/dta.1601>
41. Nguyen K.N., Saxena R., Re D.B, Yan B. Rapid L.C.-MS/MS quantification of organophosphate non-specific metabolites in hair using an alkaline extraction approach. *Et al.* 2023; 1210: 123609. Available from: <https://www.sciencedirect.com/science/article/pii/S1570023223000296>