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Investigation of partitioning of different pesticides in different water and soil/sediment systems

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Abstract

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This research focuses on simulating scenarios where surface water, containing pesticides, interacts with sediment or soil. Soil adsorption reveals insights into how pollutants interact with water and soil/sediment systems. Four pesticides with different polarities are explored for their adsorption behavior when introduced into aquatic environment. Corresponding Log$P$ values were between 3 and 5, which is characteristic for those pesticides which are present in both phases. Solutions containing pesticides were stirred with soil/sediment samples and analyzed with HPLC-UV. The distribution coefficients for adsorption (K_d), organic carbon-water partition coefficient (K_oc), and adsorption percentage at equilibrium have been calculated. PDM exhibited the highest adsorption, specifically in sediments, while TRF and FIP consistently showed high adsorption. ATC, with a low soil DT_{50}, was more present in water. Correlations indicated that pesticides with high Log$P$ values and low water solubility exhibited higher adsorption rates. Although no strict correlation was observed between soil/sediment organic matter content and adsorption properties. However, TRF, and ATC were more prevalent in higher organic soil/sediments. FIP showed no correlation with organic matter. PDM was mainly found in sediment rather than soil, possibly due to changes in mineral content and particle sizes. These pesticides exhibit instability even under colder storage conditions, emphasizing the need for immediate testing after preparation. Overall, this study unveils complex interactions between pesticides, organic matter, and storage conditions, providing valuable insights into their adsorption properties in diverse environmental matrices.

1.Introduction

Pesticides play a crucial role in modern agriculture by helping to protect crops from pests and diseases and ensuring food security and economic stability. These chemical substances are designed to target and control unwanted organisms. However, the widespread use of pesticides has raised significant concerns about their potential environmental and ecological consequences. The impact of pesticides extends beyond their intended targets, affecting non-target organisms and possibly leading to unintended effects to ecosystems. Understanding the environmental fate, persistence, and behavior of pesticide active ingredients is important to prevent harm and ensure sustainable agricultural practices. Many environmental compartments hold considerable influence over the fate of pesticides. Among these, adsorption plays an important role in the distribution and movement of pesticides within water and soil/sediment systems.

The research focuses on adsorption, a critical mechanism influencing how pesticides spread and migrate within water and soil/sediment systems. The study evaluates the Water Framework Directive's Log$P$ (K_{ow}) standard for determining the distribution of substances between sediment and water phases. According to this guideline, a substance's Log$P$ (K_{ow}) value is crucial in understanding its partitioning between sediment and water, providing meaningful insights into its behavior in the environment: below 3, it is regarded as a water pollutant, between 3 and 5 implies a potential presence in both phases, and above 5 suggests a tendency towards the sediment phase. (Voulvoulis et al., 2017)

A primary goal of this investigation is to assess the adsorption rates of various components whose Log$P$ is between 3 and 5 on soil and sediment surfaces. By examining the adsorption properties of selected pesticides in water solutions, we aim to validate the applicability of the Log$P$ (K_{ow}) criterion.

Different substances are being compared to confirm the factors influencing their final disposition. This includes evalution of soil and sediment qualities like organic matter and carbon content. These factors can change the influence of pesticide proporties attached to water. Additionally, we are assess the effects of experimental variables, such as adsorption during sample filtration, to gain a more detailed understanding of octanol-water partitioning dynamics and to avoid technical failure. In this research, a deeper understanding of pesticide behavior in the environment and more effective management strategies are being sought.

2. Literature Review

2.1 Pesticides in Environmental Phases

Pesticides are mainly applied to control unwanted plants and eliminate insects and fungi in domestic, municipal, commercial, and agricultural production systems; consequently, soil and sediments serve as the primary deposits for pesticides, influencing their widespread distribution to other environmental compartments. (Vryzas, 2018) (Allinson et al., 2016) Only a fraction of the applied pesticides reach their intended targets, and most contamination occurs through leaching, requiring natural breakdown to prevent accumulation or contamination. (Beitz et al., 1994) Due to leaching, pesticides can be found in both surface and groundwater. They are also detectable in the air, primarily bound to solid particles (aerosols) during pesticide application (Figure 1).

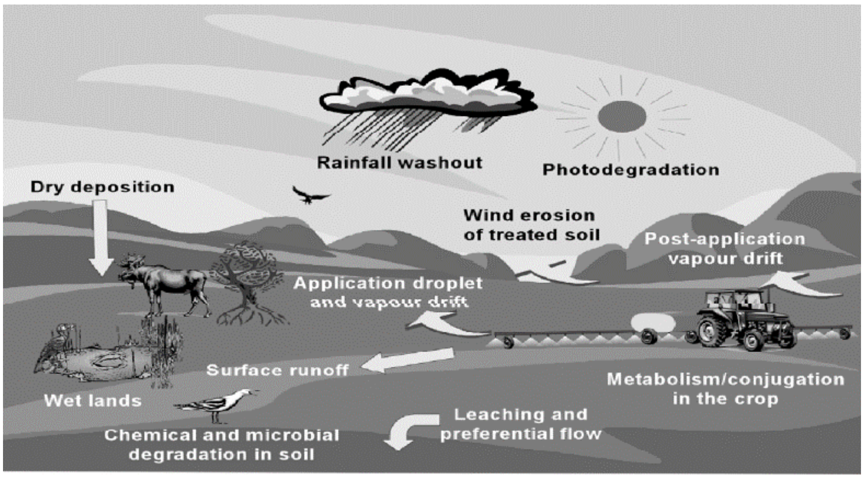

Figure 1 Routes of entry of pesticides into the atmosphere and into surface and ground waters and mechanisms of pesticide transformation in air, soil and plants (Cessna 2009).

Depending upon the particular pesticide active ingredient and specific circumstances, certain compounds have a tendency to persist within soils or sediments for extended temporal scales, spanning decades. On the contrary, different types of pesticides may disappear within a relatively short period of time, typically lasting only a few days. (Tang & Maggi, 2021) (Dimond & Owen, 1996)

In soils, pesticides enter directly or via wash-off from foliage. In addition to the inherent properties of pesticides, the soil's properties, climate, and location will interfere processes occurring in the soil or sediment to determine how pesticides will interact with other compartments (e.g surface and groundwater). (Vryzas, 2018) In addition to volatilization, processes such as adhesion to soil particles and degradation—biological, non-biological, or photodegradation—can lead to either accumulation or removal of pollutants. Due to their capacity to reduce the initial concentration of chemicals, several of these processes are considered advantageous. (Vryzas, 2018) (Kerle EA et al., 2007)

In surface water, sediments and suspended materials significantly impact how pesticides are transported. These substances act as carriers, influencing the movement and dispersion of pesticides within aquatic environments. Various pathways contribute to the introduction of pesticides into water bodies. Agricultural runoff and irrigation serve as primary conduits, transporting these chemicals from fields to surface waters. Additionally, the leaching process, where pesticides permeate through soil layers, poses a significant risk for groundwater contamination. (Vryzas, 2018) (Beitz et al., 1994) (Sharma et al., 2019) The complex process of pesticide dissipation in aquatic settings is subject to many factors. These encompass abiotic components like exposure to sunlight, fluctuations in pH levels, variations in temperature, the presence of suspended materials, and the concentration of dissolved organic matter. Chemical reactions like hydrolysis, photolysis, and the impact from aquatic organisms all play essential roles. (Vryzas, 2018) (Beitz et al., 1994) Understanding the dynamic interplay of these factors is crucial to comprehending how pesticides behave in aquatic environments (Figure 2). (Vryzas, 2018) (Kerle et al., 2007)

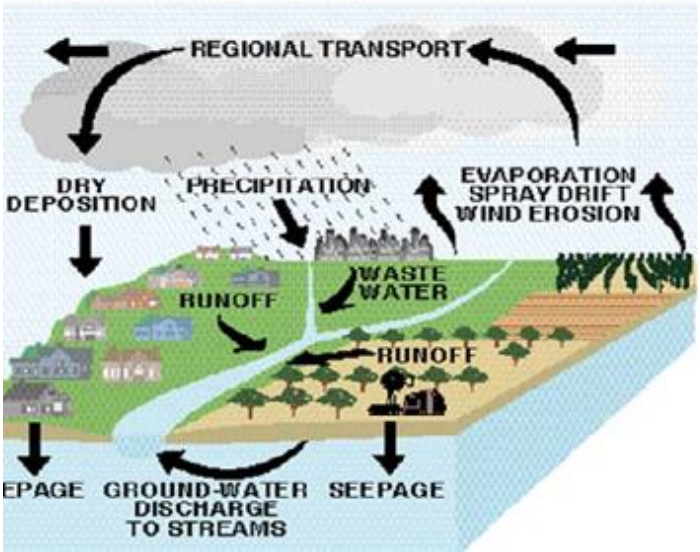

Figure 2 Pathways of pesticide into aquatic system (Kerle et al., 2007).

2.2 The EU Water Framework Directive

The European Union (EU) Water Framework Directive 2000/60/EC (WFD) introduced a comprehensive approach to water management to achieve a "good status" for water bodies throughout the EU. Unlike previous practices, the WFD emphasized the importance of catchment management and systems thinking, recognizing the complex nature of water resource management. Facilitating decentralized policy-making and encouraging public participation and adopting an experimentalist approach were crucial aspects of the planning process. (Voulvoulis et al., 2017)

Despite these ambitious goals, by 2015, nearly 60 percent of EU surface waters fell short of attaining the desired ecological status, prompting a critical examination of the WFD's effectiveness. (Voulvoulis et al., 2017) (Carvalho et al., 2019) The reasons behind the unmet expectations of the WFD include implementation challenges, narrow interpretations, ambiguities in guidance documents, limitations in transitioning monitoring networks, prioritizing compliance over understanding the broader ecosystem , and a lack of alignment between implementation practices and the directive's systemic aspirations. (Voulvoulis et al., 2017)

Carvalho emphasizes that adopting a conventional directive approach may be insufficient for improvement, highlighting the importance of acknowledging the systemic intent that underlies the Water Framework Directive (WFD). This observation implies the need to adopt a more holistic understanding and application of the WFD principles. To achieve this goal, it must use advanced monitoring approaches and various tools that have become available in recent years. There is a critical need for better integration of WFD objectives into other crucial policy domains, such as agriculture, cohesion funds, and renewable energy. The Water Framework Directive (WFD) stands as a pivotal EU initiative aimed at protecting and enhancing aquatic ecosystems while advocating for sustainable water use. (Carvalho et al., 2019)

2.3 Partition Determination

Understanding the complex behavior of pesticides in soils and sediment poses a considerable challenge. This complexity arises from the diverse range of chemicals, soil compositions, and fluctuating environmental conditions, including soil temperature and moisture levels, which wield substantial influence over pesticide retention and degradation processes. (Veronique et al., 2011)

The primary factor in the retention of pesticides in soil is the critical mechanism of adsorption (K_{op}) and the octanol-water partition coefficient (K_{ow}). These coefficients is a crucial indicator of water solubility and hydrophobicity, thus providing valuable insights into how pesticides partionate between soil or sediment and water phases in aquatic systems. Other factors, such as surface area, volume, and electronic structure, as well as the ionization and charge of pesticide molecules, are essential. (Veronique et al., 2011) (Beitz et al., 1994) In some cases model soils or sediments are used in order to study the environmental fate of pollutants in surface water. (Montuori et al., 2015) However, this thesis emphasizes octanol-water partitioning as the primary point in pursuing accurate predictions regarding pesticide behavior.

2.3.1 Octanol-Water Partition Coefficient (K_{ow})

Octanol and water are immiscible liqiuds, they form two phases upon mixing. The octanol-water partition coefficient (K_{ow}) is an indicator of a substance's hydrophobicity. (Amézqueta et al., 2020) It's defined as the ratio of a substance's concentration in octanol to its concentration in the water, measured in mass or mole per unit volume (K_{ow}=Coctanol/Cwater). Log$P$ is a dimensionless value represented the logarithm (base 10) of K_{ow}. Experimental determination involves equilibrating the substance into a water-saturated octanol phase and an octanol-saturated aqueous phase and then analyzing the phases. Solubility is temperature-dependent, especially pronounced at higher concentrations—additionally and pH impact solubility. Handling volatile substances demands caution. (Guidance for reporting on the environmental fate , 2023) (Kozerski et al., 2014). Factor, like time does not influence the equilibrium, however worthy of note that it takes time to reach the equilibrium. Fresh loads to surface water may result in higher levels dissolved in the water phase, thus actual concentration may differ from the equilibrium values. (He *et al.*, 2016b) (Sharma et al., 2019) (Shah & Parveen, 2023)

As K_{ow} is inversely correlated with water solubility, it can be used to predict the rate of adsorption. The value of K_{ow} depends on a compound's polarity and hydrophobic characteristics. According to the WDF, ompounds with K_{ow} values below 3 tend to reside in the water phase, while those above 5 are lipophilic and prefer nonpolar phases like octanol or sediment. Values between 3 and 5 indicate a presence in both phases (Figure 3). (Mörtl, 2023)

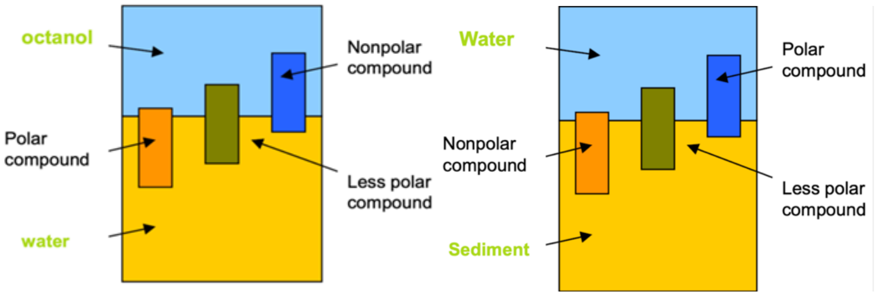

Figure 3. Distribution of compounds with different polarities in water and octanol/sediment systems (Mörtl, 2023)

In Figure 4, the transport process in a chromatographic system, when there is no equilibrium. The two compounds exhibit two different rates of movement, or mobilities, due to the constant flow of the mobile phase and partition between mobile and stationary phase. The process is similar to the river/sediment system. Compound (k2), which possesses a high Log*P* constant, tends to be retained by the sediment phase. In contrast, compounds like (k1) with lower values tend to move along with the water phase, and any suspended particles in the river phase further assist in their transport.

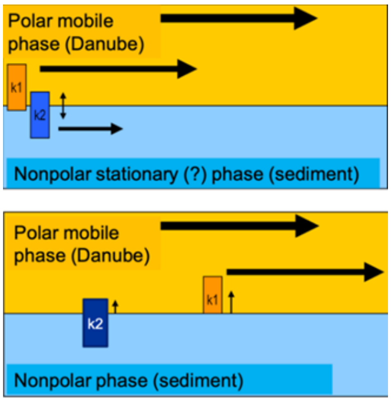

Figure 4. Different mobilities of two compounds in water-sediment system (Mörtl, 2023)

K_{ow} values have significant effect to pesticide uptake rates by the plants are well. Those with a high Log*P* (> 4) are generally non-systemic, contact pesticide active ingredients. In contrast, plants absorb and then transpor moderately less lipophilic ones via the xylem to developing shoots (typical LogP values are between 0.5 and 3.5). It's important that roots exhibit better absorption capabilities for more lipophilic insecticides. For example, neonicotinoids, a widely utilized class of insecticides, tend to possess low LogP values, allowing for effective systemic control of specific insects. (Schriever, 2020) The transpiration stream concentration factor (TSCF) quantitatively characterizes the extent of root uptake. It is defined as the ratio of a compound's concentration in the transpiration stream (xylem) to its concentration in the external solution. Figure 5 illustrates that even subtle differences in lipophilicity can significantly affect root uptake. (Jeschke et al., 2013) (Schriever, 2020)

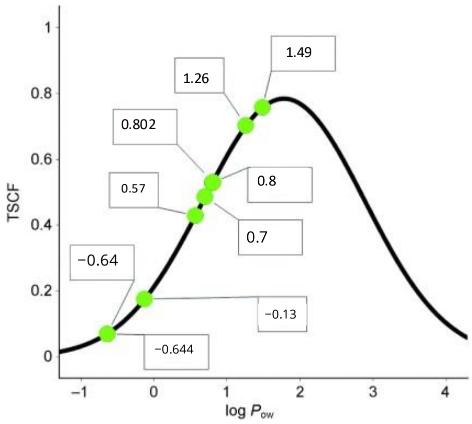

Figure 5 The relationship between Transpiration Stream Concentration Factors (TSCF) of nAChR agonists and their corresponding log K_{ow} values (Jeschke et al., 2013)

2.3.2 Soil Adsorption

Adsorption in soils is a crucial process where substances adhere to soil particles to maintain surface equilibrium. Soil particles can retain various substances, including soil components, essential plant nutrients, and harmful environmental contaminants. These substances may be positively charged cations, negatively charged anions, or nonionic molecules. Soil properties and environmental factors also influence this interaction. (Huang, 1980)

Additionally, adsorption affects soil elements like enzymes, microorganisms, and plant roots. Understanding these processes is crucial for mitigating effects on pesticide pollution on the soil health and minimizing risks from environmental pollutants. Furthermore, adsorption mainly influences the behavior of pesticides within soil, sediment, and water systems. (Huang, 1980)

There are a few ways to calculate adsorption in soil. K_{oc}, K_d, or K_f measure how easily a substance moves in soil. K_{oc}, the organic-carbon partition coefficient, can be predicted using the octanol-water partition coefficient (K_{ow}) and water solubility, and it indicates how strongly a substance binds to organic matter in soil. A higher K_{oc} value means a stronger attachment, limiting the substance's movement in the soil. Conversely, a low value suggests that the substance can move more freely in the soil. K_d, the distribution coefficient, indicates how strongly a pesticide attaches to soil particles. A higher K_d value suggests lower chances of leaching or causing runoff. Furthermore, K_f, the Freundlich adsorption coefficient, can be used to calculate soil adsorption to a certain extent, and it helps understand how substances adhere to soil. It shows the relationship between a substance's concentration in the soil and the soil solution. (OECD/OCDE. Test No. 106: Adsorption , 2000) (Kodešová et al., 2011) (Sparks, 2003) (Kerle EA et al., 2007) (see below)

In this thesis, the formulas from 1 to 5 are applied in soil adsorption experiments. In addition, Table 1 provides definitions of each symbol and its units. Furthermore, Table 1 provides detailed definitions for each symbol and its respective units. The first equation illustrates the percentage of adsorption at a given time point (t_i) by dividing the quantity adsorbed onto the solid phase (as described in equation 2) at that specific time point (t_i) by the initial amount applied. The adsorption percentage at equilibrium (as shown in equation 3) is calculated by dividing the amount adsorbed onto the solid phase at equilibrium by the initial applied quantity. Equation 4 shows concentration of two distinct methods for calculating the distribution coefficient at adsorption equilibrium, denoted as K_d. Afterward, the organic carbon-water partition coefficient, K_{oc}, is derived from K_d and organic carbon using equation 5. (Guidance for reporting on the environmental fate , 2023) (OECD/OCDE. Test No. 106: Adsorption , 2000) (OECD/OCDE. Test No. 117: Partition Coefficient, 2014) Units are in Appendix A.

1. $A_{t_i}(\%) = \frac{m_s^{ads}(t_i)\times 100}{m_o}$
2. $m_s^{ads}(t_i) = m_o - m_{aq}(t_i)$
3. $A_{eq}(\%) = \frac{m_s^{ads}(eq)}{m_o} \times 100\%$
4. $K_d = \frac{A_{eq}}{100-A_{eq}} \times \frac{V_o}{m_{soil}} = \frac{C_s^{ads}(eq)}{C_{aq}^{ads}(eq)} = \frac{m_s^{ads}(eq)}{m_{aq}^{ads}(eq)} \times \frac{V_o}{m_{soil}} \, (cm^3 g^{-1})$
5. $K_{oc} = K_d \times \frac{100}{\%oc} \, (cm^3 g^{-1})$

2.3.2.1 Soil Properties' and Environmental Conditions Impact on Pesticides

The properties of soil and environmental factors have a significant impact on pesticides. These include water content, temperature, pH, carbon content, and other relevant properties—the information on these factors is provided in Table 1. (Beitz et al., 1994) (Veronique et al., 2011) In the laboratory, various parameters such as soil texture, moisture content, pH level, organic matter (OM), nutrient levels, cation exchange capacity (CEC), electrical conductivity (EC), bulk density, and permeability are measured. On-site recorded parametr yield a broader range of information and may also contribute to better evaluation. This includes visual assessments of soil color, structure, and topography. Moreover, it is important to measure and evaluate soil temperature, microbial activity, vegetation cover, and land use practices (past and present).

Table 1 Soil properties and environmental conditions that influence environmental fate of pesticides (Veronique et al., 2011)

Soil properties:	
Minerals	Such as clays, oxides, and hydroxides play a significant role in pesticide adsorption. Clays are mainly hydrophilic, and adsorption occurs on external surfaces. Oxides and hydroxides have high surface activity.
Organic matter	Organic matter is a significant sorbent of pesticides in soil due to its chemical reactivity. The organic matter directly impacts the water retention capability and adsorption tendencies (OM) content. Soils characterized by a higher OM content will retain water for an extended period.
Soil pH	Soil pH affects the adsorption of ionic pesticides. Depending on the charge of the pesticide, adsorption increases or decreases with pH.
Soil structure	Soil structure is characterized by bulk density and pore geometry, which affects pesticide movement. No-till soils and high organic matter content, generally have higher pesticide retention. Soils with high permeability are easy to leaching, whereas those with low permeability reduce the pace of downward pesticide movement.
Effects of environmental conditions	
Water content	It influences the specific exchange surface between solid and liquid phases. Adsorption increases with higher water content.
Temperature	Generally, adsorption decreases at higher temperatures. Fast sorption increases with decreasing temperature, while slow sorption is more rapid at higher temperatures.
Spatio-temporal variability of retention:	
Spatial variability	It can be laterally or vertically. Variation in organic carbon content is a major contributing factor.
Temporal variability	It is influenced by residence time in the soil. Over time, interactions evolve to form non-extractable residues, which persist in the soil.

2.3.3 Distinction between Soil and Sediment

Within this study, it is important to create a distinction between soil and sediment. Soil, constituting the uppermost layer of the Earth's surface, is a complex combination of mineral particles, organic matter, water, and air. It plays an essential part in sustaining plant life and supporting diverse ecological communities. In contrast, sediment comprises loose, solid particles that settle at the bottoms of water bodies, originating from erosion and weathering. The geological and environmental contexts in which these materials form are considerably different. (Herz & Garrison, 1998)

Long-term geological, climatic, and biological influences on soil formation result in a diverse matrix with varying levels of mineral content, organic material, and physical structure. Sediment, which, primarily arises from the erosion of existing geological formations, undergoes transport and deposition through aqueous, aeolian, or glacial mechanisms. From a compositional perspective, soil consists of a range of components, which include mineral particles categorized as sand, silt, and clay, as well as organic matter, water, and air. This composition is flexible and susceptible to modification by factors such as climate, vegetation cover, and parent material. (Herz & Garrison, 1998) (Banning, 2002) Please refer to Figure 6 for an illustration of the properties and conditions that influence the fa te of pesticides in the soil environment.

In contrast, sediment predominantly comprises mineral constituents, with the presence of organic matter dependent upon the source and location. In addition, it is important to note that soil and sediment possess a wide range of chemical and physical characteristics that play a crucial role in the adsorption processes of pesticides. Soils exhibit a range of attributes, such as pH levels, cation exchange capacity (CEC), and textural features, which combined impact their ability to adsorb pesticides. The properties of sediment, on the other hand, depend on the material it comes from and the processes that move it, giving it different absorptive capacities. (see section 2.3.2.1) (He *et al.*, 2016b) (Rasool et al., 2022)

Pesticides are frequently monitored in surface water, but only some studies have focused on investigating pesticide residues in water and sediment samples. In comparison to water samples, sediment samples generally exhibit less temporal variability in pesticide concentration. Pesticide concentrations in sediment are less affected by extreme rainfall and runoff than in water samples. (Vryzas, 2018) (Shah & Parveen, 2023b)

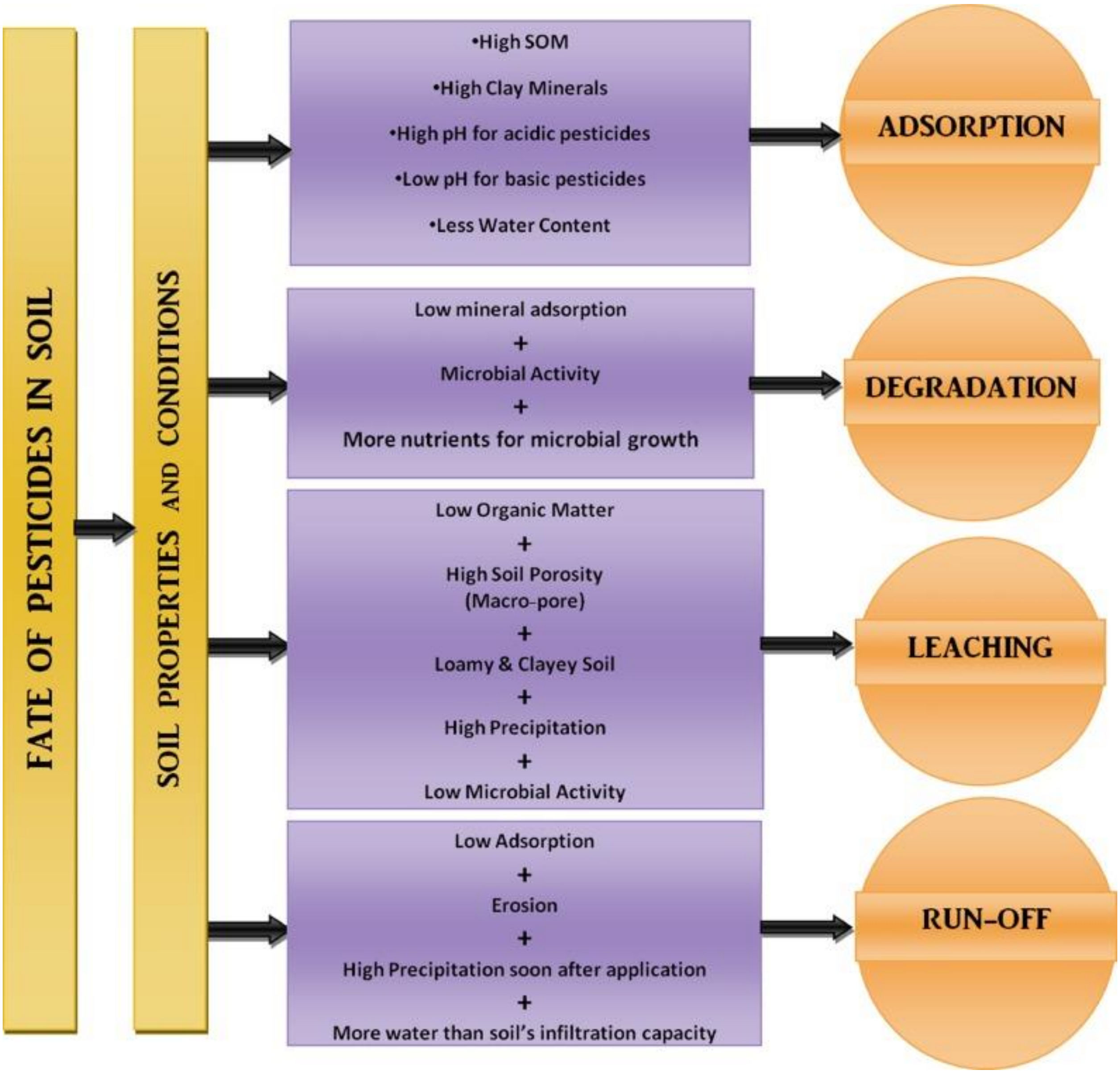

Figure 6 Properties and conditions determining the fate of pesticides in soil environment. (Rasool et al., 2022)

2.4 General characteristics of pesticides used as model compounds with different Log P values

This investigation targeted five pesticides: propiconazole (PCZ), acetochlor (ATC), fipronil (FIP), trifluralin (TRF), and pendimethalin (PDM). (See Figture 7) These particular pesticides were selected due to their log K_{ow} values falling between 3 and 5 in Table 2, which is indicative of their presence in both soil and water phases. The primary aim of this study was to evaluate the behavior and distribution of these compound in various soil/ sediment, and water phases.

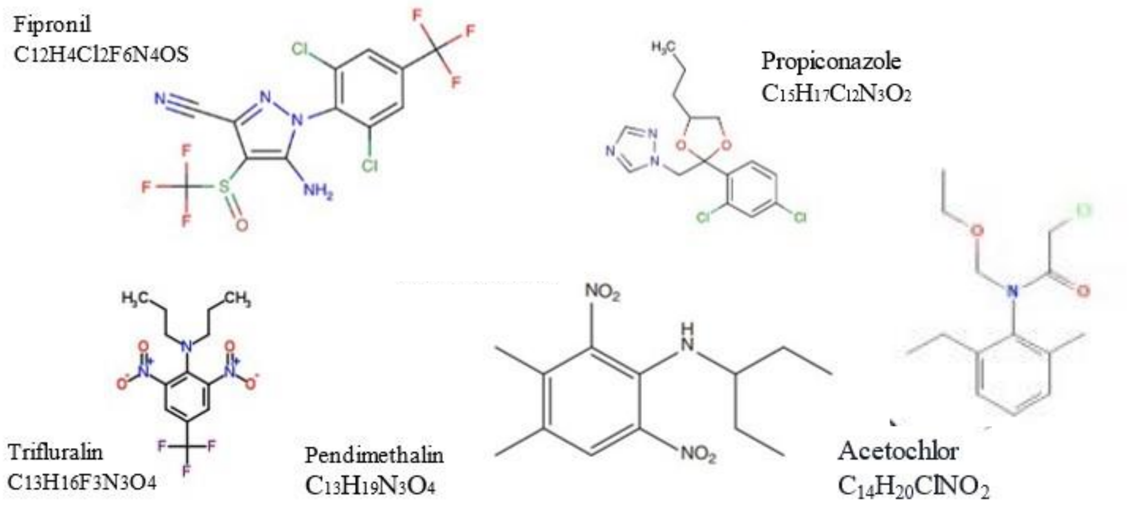

Figure 7. 2D chemical structure of pesticides studied in the second investigation

PCZ is a widely used fungicide in agriculture, turf management, and wood treatment, effective against various crops, including mushrooms, corn, wild rice, peanuts, almonds, sorghum, oats, and pecans, as well as fruits like apricots, plums, prunes, peaches, and nectarines. It belongs to the chloroacetanilide class of herbicides, and it was developed by Janssen Pharmaceutical in 1979. Its appearance can range from a clear to pale yellow viscous liquid, depending on the isomer ratio. It is often supplied as emulsifiable concentrates or wettable powder. The material remains stable until it reaches 320. (Gad and Pham, 2014) (University of Hertfordshire, n.d)

The Fungicide Resistance Action Committee (FRAC) is an international organization that provides guidelines and standards for classifying fungicide resistance in plant pathogens and categorizes different modes of action of fungicides based on their biochemical and physiological effects on target pathogens. This helps in managing and preventing the development of resistance in agricultural settings. The FRAC code consists of a combination of numbers and letters that represent specific groups of fungicides that comprise three key components. First, a numerical identifier represents the mode of action. For instance, Group 3 pertains to demethylation inhibitors (DMIs) or sterol biosynthesis inhibitors (SBIs), which disrupt the ergosterol biosynthesis pathway in fungi. Second, a letter designates the chemical group to distinguish various chemical structures within a particular mode of action group. For example, within the DMI (Group 3) classification, there are distinct chemical groups like triazoles (e.g., PCZ). Lastly, a specific chemical may be appended to the FRAC code to precisely identify a particular fungicide. This classification system aids in informed decision-making for fungicide selection and rotation, critical in managing resistance in agricultural settings. (FRAC, n.d.) Therefore, PCZ falls under category 3. PCZ's mode of action involves demethylating C-14 during ergosterol biosynthesis, accumulating C-14 methyl sterols. The reduced production of ergosterols hampers fungal growth, as they are vital for the structure of fungal cells. This effectively stops or hinders further infection and invasion of host tissues. Despite being banned in the UK and EU, PCZ is extensively utilized worldwide, particularly in the Asia-Pacific region, where it accounts for nearly half of the market share of PCZ. It can be detected through gas chromatography (GS). (MacBean C, 2012) (PCZ Market, n.d)

ATC, an herbicide belonging to the chloroacetamide class, is primarily applied to corn, soybeans, and sugar beets. It was introduced in 1985 and has seen widespread use in the USA. However, it was prohibited in the EU in 2011 due to its toxicity to aquatic organisms, birds, and human health. ATC exists in a pale-yellow liquid state and is most effective before germination begins. It effectively controls most annual grassy and broadleaf weeds, though its efficacy against perennial weeds is limited. Often, it is used in combination with other herbicides to achieve broad-spectrum weed control. The substance remains stable for over two years at 20 degrees Celsius. (Rajinder, Mann, p. 2021) (Acetochlor - General Information, n.d.) (University of Hertfordshire, n.d.-b)

The HRAC (Herbicide Resistance Action Committee) and WSSA (Weed Science Society of America) are organizations that have provided guidelines and standards for the classification of herbicide resistance in weeds and are similar to the FRAC system for fungicides. (HRAC MOA 2020 Revision Description and Master Herbicide List, p. 2020) The HRAC and WSSA classifications both utilize a system comprising numbers and letters to denote specific groups of herbicides. The HRAC system employs a Mode of Action Group (MOA) identified by a numerical code to elucidate the overarching mechanism of action, similar to FRAC. In contrast, the WSSA classification system uses letters to represent the general mode of action and then a number to specify the specific site of action. For instance, the WSSA Code B2 pertains to ALS inhibitors, targeting the site of action—acetolactate synthase (ALS) inhibitors. (Herbicides, n.d) ATC is in the HRAC and WSSA groups; it is labeled as K3, 15, which shows that it stops the production of very long-chain fatty acids during cell division and mitosis. This disruption leads to interrupted seedling growth and the possibility of abnormal development. ATC can be detected through HPLC and GS-MS. (MacBean C, 2012)

Scientists from Rhone-Poulenc discovered and commercialized FIP, a popular non-systemic, chiral phenyl-pyrazole insecticide, in the UK in 1987. It was known under various trade names like Frontline, Combat, and Max-force. This insecticide is applied in agricultural, nonagricultural, and residential settings to combat pests, including fleas, ticks, ants, beetles, cockroaches, and termites, offering broad-spectrum activity. While it is used in some EU countries, such as the Netherlands, Belgium, and Hungary, it sees more widespread application outside the EU, particularly in Australia, the USA, and Iran. Fipronil is typically found in fine granules, appearing as a white to beige-colored powder, and is dissolved in water for use. Stable to heat but slowly degrades in sunlight. (University of Hertfordshire, n.d.-b) (Fent, 2014) (*Worldwide development of fipronil insecticide*, n d)

The Insecticide Resistance Action Committee (IRAC) Mode of Action Classification is a systematic framework used for the classification of insecticides, similar to FRAC and HRAC. The IRAC classification system employs a distinctive code structure, encompassing two integral components: the Mode of Action Group and the Chemical Group. (Insecticide Resistance Action Committee, 2023) In the IRAC Mode of Action classification scheme, FIP belongs to the 2B group. It acts on insect nervous systems by noncompetitively blocking specific channels (the GABA-gated chloride channel), displaying high selectivity for insects. It has a low to moderate level of toxicity for mammals. It can be detected through high-performance liquid chromatography (HPLC) with UV detection, as well as gas chromatography and mass spectrometry (GC-MS). (MacBean C, 2012)

Trifluralin is a widely used dinitroaniline herbicide for preemergence control of many annual grasses and broad-leaved weeds, including cotton, soybeans, oilseed rape, sunflowers, and peanuts. For some plants, it can be used post-planting. The physical state is an orange-yellow crystalline solid. It was initially approved in 1961 and remains a common herbicide in the United States and Australia. In 2009, it was banned in the EU due to concerns about its persistence in soil and groundwater. (University of Hertfordshire, n.d.-b) (MacBean C, 2012) In the herbicide resistance classification (HRAC, WSSA), it falls under the K1 and 3 categories, which belong to "inhibition of microtubule assembly". The dinitroaniline class selectively hinders the production of microtubules in constructing cell walls (inhibition of mitosis and cell division). This leads to a suspension in root development and, ultimately, the death of the targeted plant. (Wallace, 2014) Detection and quantification of TRF levels can be achieved using high-performance liquid chromatography or gas chromatography/mass spectrometry. (MacBean C, 2012)

The American Cyanamid Company created PDM, also referred to by its trade name "Prowl," in the early 1970s. PDM is a synthetic herbicide widely utilized to control broadleaf weeds and woody plants in crops like corn, rice, cotton, and potatoes. Available in various formulations, it can be applied before or after seed emergence. The physical state is orange-yellow crystalline. PDM is available in different formulations, including liquid, solid, granule, powder, and emulsifiable concentrate. (Ramasahayam, 2014) (University of Hertfordshire, n.d.-b) The substance exhibits high stability in storage, withstanding temperatures above 5 degrees Celsius and below 130 degrees Celsius. It is non-flammable and non-explosive. It is utilized globally, including in EU member states, the USA, and Australia. According to the Herbicide Resistance Action Committee (HRAC), it falls under the classification K1, while in the Weed Science Society of America (WSSA), it is categorized as 3. Consequently, its mode of action aligns with that of TRF, as it functions by inhibiting mitosis and cell division. Additionally, it can be detected through gas chromatography (GS). (MacBean C, 2012)

2.4.1 Environmental fate and toxicity

PCZ, with a Log*P* value of 3.72 at pH 7 and 20°C, demonstrates moderate solubility in water and relatively low vapor pressure. The compound is moderately persistent in terrestrial environments, primarily undergoing biotransformation. It is also moderately persistent in aquatic environments and may contaminate ecosystems through runoff under heavy rainfall as it has a low GUS leaching potential index. Exposure to PCZ can occur through dermal contact, inhalation, and ingestion. PCZ is listed as a possible human carcinogen (Group C), but this assessment is based on a study with excessive toxicity in high-dose groups. Ecotoxicologically, PCZ can be toxic to fish and *Daphnia magna* (see Table 3), while it has relatively low toxicity to birds and bees. The dioxolane ring and the propyl side chain are both hydroxylated, which is the main mechanism of degradation. 1,2,4-triazole is finally produced as a result of these activities. (Gad and Pham, 2014) (University of Hertfordshire, n.d.-b) (MacBean C, 2012)

FIP has low solubility in water and is not easily vaporized. FIP exhibits moderate persistence, degrading through processes like photodegradation and hydrolysis. It has low soil mobility and limited potential for groundwater contamination due to a low leaching index of 2.06. Exposure to fipronil can occur dermally, through inhalation, or orally. It is classified as a possible human carcinogen (Group C). In ecotoxicology, fipronil exhibits variable toxicity to non-target species, being highly toxic to fish, aquatic invertebrates, bees, and some birds but nearly nontoxic to certain waterfowl, earthworms, soil microorganisms, and aquatic plants. This substance is considered non-mutagenic and non-teratogenic, and it does not have adverse effects on reproductive performance. While it is highly toxic to bees upon direct contact, it can be safely used as a soil or seed treatment. In terms of degradation, major pathways in aerobic soil conditions lead to the formation of sulfone and amide compounds. In anaerobic conditions, degradation primarily results in sulfide and amide compounds. (Fent, 2014) (University of Hertfordshire, n.d.-b) (MacBean C, 2012) See Table 2 for other parameters.

Table 2 Parameters related to the environmental fate of the selected pesticides (University of Hertfordshire, n.d)

Name	Log*P* at pH7, 20 °C	Water solubility (mg/L)	Pesticide type	K_d (mL/g) range	K_{oc} (mL/g) Linear	Vapour pressure at 20°C (mPa)	SD DT_{50} (typical) days	WS DT_{50} days	WP DT_{50} days	GUS leaching potential index
PCZ	3.72	150	Fungicide	33.7[a]	382-1789 [a]	0.056	71.8	561	6	1.58
FIP	4.00	3.78	Insecticide	5.34 to 13.85[b]	353-6963 [b]	0.002	142	68	54	2.06
ATC	4.14	282	Herbicide	0.13-17.0[a]	28-377[a]	$2.2*10^{-02}$	14	19.7	40.5	1.67
TRF	4.83	0.221	Herbicide	105-30903[b]	3900-15800[b]	9.5	133.7	5.5	13	0.13
PDM	5.2	0.33	Herbicide	120-677[a]	10241-36604[a]	3.34	182.3	16	4	-0.28

[a] -Verified data used for regulatory purposes

[b] - Peer reviewed scientific publications

SD DT50 (Soil Dissipation Time at Half) refers to the dissipation time at half in the soil. It represents the time it takes for half of the initial concentration of a pesticide to degrade or dissipate in the soil environment.

WS DT50 (Water-Sediment Dissipation Time at Half) stands for the Dissipation Time at Half in both water and sediment. This parameter indicates the time it takes for half of the initial concentration of a pesticide to degrade in a combined water and sediment environment.

WP DT50 (Water-Plant Dissipation Time at Half) represents the dissipation time at half in both water and plants. This parameter signifies the time it takes for half of the initial concentration of a pesticide to degrade in a combined water and plant environment.

The units of measurement for these parameters are expressed in days.

ATC exhibits moderate solubility in water and is not easily vaporized. It possesses moderate mobility in soil and does not persist for extended periods. With a Log*P* value falling between 3 and 5, it is classified as a potential pollutant in water and soil environments. However, its low leaching index of 1.67 suggests a minimal risk of groundwater contamination, while surface runoff poses a higher potential for pollution. In terms of its half-life, ATC remains stable in aquatic systems even frequently found in surface water of Hungary. 32. (Mörtl M, 2021) In a water-sediment system, it undergoes rapid degradation. Considering its chemical attributes, the likelihood of leaching into groundwater is anticipated to be low. Furthermore, ATC shows resilience to both aqueous photolysis and hydrolysis at pH 7 and 20°C. (University of Hertfordshire, n.d) The main metabolites are water-soluble acids that are produced via glutathione conjugation or by the oxidative displacement of chlorine. These substances are then metabolized to produce acids that contain sulfur, such as sulfonic and sulfinylacetic acids. (MacBean C, 2012)

TRF exhibits relatively low mobility and persists in soil. It has been frequently detected in soil of Hungary even after it's ban (Mörtl M, 2021). It has a low leaching potential index of 0.12. Although TRF is practically insoluble in water, it is lipophilic (with a high Log*P*) and moderately volatile, which means it can be detected in soil and sediments near water bodies. Exposure to TRF can occur through inhalation of contaminated air, absorption through the skin, or ingestion of contaminated plants or animals. Additionally, TRF remains stable in aqueous hydrolysis at 20°C and pH 7, but degrades rapidly in aqueous photolysis at the same pH. TRF is highly toxic to fish, while it exhibits low toxicity to mammals. (Wallace, 2014) (University of Hertfordshire, n.d) The process of metabolism includes dealkylation of the amino group, nitro group conversion to amino group, trifluoromethyl group partial oxidation to carboxy group, and breakdown into smaller components. (MacBean C, 2012)

PDM's solubility in water is low, and it is primarily soluble in organic solvents. PDM tends to bind to soil and sediments, reducing its leaching potential. Therefore, it has a low potential for leaching into groundwater. In the atmosphere, it degrades through reactions with hydroxyl radicals. While slightly toxic through ingestion, it is non-irritant to the skin. Although not considered mutagenic, high doses in rats led to thyroid follicular adenoma, prompting a potential human carcinogenic classification. Regarding ecotoxicology, PDM poses a high risk to freshwater fish and aquatic invertebrates and can be detrimental to aquatic plant species. It's slightly toxic to birds. (Ramasahayam, 2014) In soil, the degradation of PDM involves the oxidation of the 4-methyl group on the benzene ring, leading to the formation of carboxylic acid through an intermediate alcohol. Additionally, the amino acid nitrogen is subject to oxidation in this process. The toxicity values of these pesticides to non-target organisms are provided in the Table 3. (University of Hertfordshire, n.d) (MacBean C, 2012)

Table 3 Toxicity values of individual pesticides for test species (University of Hertfordshire, n.d)

Names	Aquatic invertebrates		Fish		Mammals		Honeybees	
	*Daphnia Magna		*Oncorhynchus mykiss		*Rats		*Apis mellifera Acute LD$_{50}$ (g/bee)	
	Acute 48h EC$_{50}$ (mg/L)	Chronic 21d NOEC (mg/L)	Acute 96h LC$_{50}$ (mg/L)	Chronic 21d NOEL (mg/L)	Acute oral LD$_{50}$ (mg/kg)	Short term dietary NOEL (mg/kg)	Contact	Oral
PCZ	10.2^{m}	0.31^{m}	^{b}2.6^{m}	0.068^{m}	550^{m}	2.7^{h}	> 100^{l}	> 100^{l}
FIP	0.19^{m}	0.068^{m}	0.248^{m}	0.015^{m}	92^{h}	0.35^{h}	0.0059^{h}	0.00417^{h}
ATC	8.3^{m}	0.022^{m}	0.36^{m}	0.13^{m}	1929^{m}	> 10^{h}	> 200^{l}	> 100^{l}
TRF	0.245^{m}	0.051^{m}	0.088^{h}	^{a}10^{m}	> 5000^{l}	2.4^{h}	> 100^{l}	> 100^{l}
PDM	0.147^{m}	0.0145^{m}	0.196^{m}	^{a}0.006^{h}	4665^{l}	> 500^{l}	100^{m}	> 101.2^{l}

^{h}*high,* ^{m}*moderate,* ^{l}*low,* ^{a} *Pimephales promelas,* ^{b} *Leiostomus xanthurus,*

EC$_{50}$ (Effective Concentration 50%) is the amount of a substance that is needed to have a certain effect on a living system.

LC$_{50}$ (Lethal Concentration 50%) is the concentration of a substance causing mortality in 50% of a test population.

LD$_{50}$ (Lethal Dose 50%) signifies the dose of a substance resulting in mortality in 50% of a test population within a specified period.

NOEL (No Observed Effect Level) is refers to the highest dose or level of exposure in a controlled study where no adverse effects or observable effects are detected.

According to the World Health Organization's recommended classification of pesticides by hazard in 2020, which is based on LD50 for rats, their toxicities towards mammals vary. ATC is classified as slightly hazardous (Class III), whereas FIP, PDM, and PCZ are considered moderately hazardous (Class II). TRF is unlikely to pose an acute hazard (Class U). (Fent, 2014) (Ramasahayam, 2014) (Gad and Pham, 2014) (Wallace, 2014)

2.5 Analytical method for pesticide analysis in soil and water

A different instruments are available for the detection and measurement of pesticides. For example, the gas chromatograph (GC) separates complex mixtures of compounds to accurately identify and quantify pesticide residues. It does this by vaporizing and injecting a sample into a column, where it interacts with a stationary phase, allowing for precise separation based on compound characteristics. The liquid chromatograph (LC) serves a similar purpose to the GC but uses a liquid mobile phase instead of carrier gas. This method is particularly effective for compounds with lower volatility and thermally labile. Mass spectrometry (MS) coupled both GC and LC by determining compound identities based on their mass spectra. By measuring the charge ratio of ions formed from the sample, MS allows precise identification of specific pesticides, enhancing the accuracy and reliability of pesticide analysis. (Engelhardt, p. 1989) Instruments like the Ultraviolet-Visible Spectrophotometer (UV-Vis) measure the absorption of light by molecules in a sample, providing information about their concentration. While it may not offer compound-specific identification, UV-Vis Spectrophotometry serves as an initial, cost-effective screening tool for pesticide residues. However using DAD detection mode provide additional information,which can help to exclude false positive result or detect interfering compound (Rocha et al., 2018)

An immunological test called enzyme-linked immunosorbent assay (ELISA) can find and measure certain pesticides by letting antigens (pesticide molecules) interact with antibodies. Although it only provide information on single compound, ELISA offers a rapid and cost-effective means for screening large numbers of samples. (Alhajj, 2023) (Engvall, 2010)

Before analysis, the pesticide sample is prepared using the QuEChERS method nowadays. This dispersive solid-phase extraction method (d-SPE) consists of an extraction phase, a clean-up phase, and analysis. The quick, easy, cheap, effective, rugged, and safe method is considered one of the most effective extraction methods. Even though it is easier, some compounds are better prepared by using the liqiud-to-liqiud method. It was initially invented to recover pesticide residue from vegetables. (González-Curbelo et al, 2015) (QuEChERS: About the method, n.d) However, in this thesis, HPLC is used.

2.5.1 High-Performance Liquid Chromatography (HPLC)

Liquid chromatography (LC) was first discovered in the early twentieth century to separate colored compounds. To measure volatile compounds, gas chromatography is used, whereas for less volatile or thermally labeled compounds, LC is the method of choice. (Poole, 2020) HPLC was developed to address the limitations of conventional LC. HPLC separation relies on the interactions between chemical compounds and the stationary/mobile phase system. This technique separates a sample into its individual components based on how strongly they bind to the mobile and stationary phases. HPLC serves both qualitative and quantitative purposes, identifying and quantifying compounds. Instruments usually consist of solvent delivery pumps, a sample injector, a column oven, a detector, and a workstation. (Douglas A et al, 2017)

The solvent is the mobile phase, typically a mixture of polar and non-polar liquids. Its composition varies based on the sample, stationary phase (column), and chromatographic conditions (temperature and flow rate), but it must be free of dissolved gases and particulates to prevent column blockage and ensure consistent results. The mobile phase composition (the ratio of polar to non-polar components) is crucial for the efficient separation. The goal is to elute all compounds quickly while maintaining peak resolution. Retention time is the duration for a substance to pass through the chromatographic column and reach the detector, serving as an identifier. The peak area represents the space of a specific peak on the chromatogram, proportionally reflecting the substance's quantity in the sample. (Sadaphal & Dhamak, 2022)

Within the column, sample components interact with the packing material. Stronger interactions with the stationary phase lead to longer retention times. Columns packed with solid materials like silica or alumina and their surface is chemically modified by interacting groups. (Serban et al, 2013)

HPLC uses different stationary phases. In normal phase LC, the stationary phase is more polar than the mobile phase, while in reverse phase, it is less polar. In reverse-phase LC, compounds with lower polarity have longer retention times. Normal phase columns use alumina or silica packing, while the reverse phase utilizes alkyl (aliphatic) or phenyl-bonded (modified) phases. Reverse-phase HPLC is widely used, except for specific cases like inorganic ions, polysaccharides, polynucleotides, and highly hydrophobic compounds, where it is less effective. (Biosciences, 1997) An isocratic elution occurs when the mobile phase's composition remains constant throughout the HPLC separation. Gradient elution involves changing the mobile phase composition during the run. It is preferred for samples with a wide range of polarities, providing comprehensive peak separation. (Sadaphal & Dhamak, 2022)

The detector, positioned at the column's end, provides detector responce (signal) for sample components without detecting the solvent. Various detectors are used to specific applications. UV absorption detectors are prevalent, especially for medium- to large molecules. Furthermore, mass spectrometry is also often used. Fluorescence and refractive index detectors are applied in specialized cases. (Douglas A et al, 2017) (Serban et al, 2013)

3 Materials and methods

3.1 Soils and sediments

There were five types of soil and five different sediments. All soil and sediment samples were taken from the soil's upper layer (0–10 cm) and air-dried. Additionally, the samples' colors were analyzed from the Munsell color chart (Munsell colors , 2016) (refer to Table 4). Soils and sediments listed from 1 to 7 in Table 4 were all gathered in Hungary on October 10, 2020 (see Figure 8). The sediment labeled with laboratory code 1419 was procured from the Séd stream near to Veszprém, Hungary, on August 13, 2020, specifically from the northern area of the Nádor channel. The two soils, HOC and LOC, as mentioned in Table 4, were obtained from agricultural fields near the Research Farm of the University of Natural Resources and Life Sciences (BOKU) in Vienna on April 12, 2018. The abbreviations HOC and LOC mean higher and lower organic content, respectively.

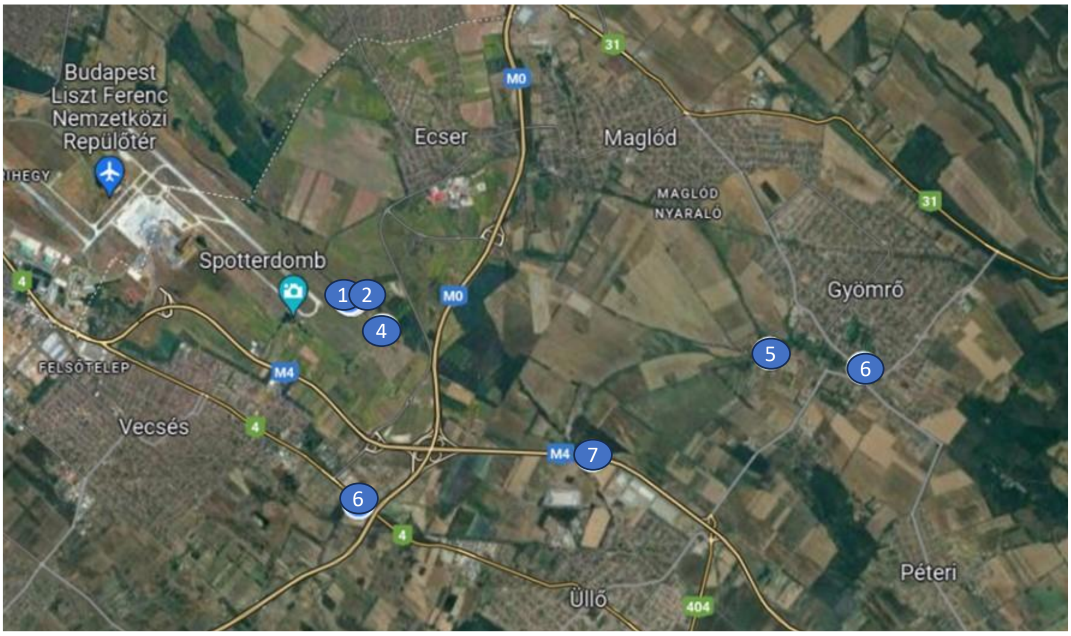

Figure 8 Sampling sites for soils and sediments according to GPS coordinates

Table 4. Detailed information on all the soils and sediments

Code	Lab Code	Type, color	% OC on dry matter	Location	Location details
1	1426	Dark yellowish brown soil 3/6	2.94^{L}	Ecser 2233	Agricultural field close to channel 172

2	1425	Light yellowish brown soil 6/4	3.04^{L}	Monori	Close to channel 172
3	1424	Brown soil 4/3	17.87^{H}	Gyömrő 2230	Near to Tözeges lake
4	1427	Brownish yellow sediment 6/8	2.37^{L}	Vecsés 2220	Intersection of 2 channels: 172, Maglódi 17
5	1421	Dark yellowish brown sediment 4/4	14.83^{H}	Gyömrő 2230	Halas 18 csatorna stream
6	1423	Yellowish brown sediment 5/8	5.75^{L}	Üllő 2225	Intersection of 2 channels: Gyáli 1, Maglódi 17
7	1420	Brownish yellow sediment 6/8	16.05^{L}	Soroksar, 1420	Soroksár, Ráckeve Duna
8	1419	Brownish yellow sediment 6/6	15.31^{H}	North of the Nádor channel	Séd stream in Veszprém
9	HOC	Dark yellowish brown soil 4/4	4.1^{L}	N48.1833 E16.55	Agricultural field close to BOKU, Vienna
10	LOC	Light yellowish brown soil 6/4	3.0^{L}	N48.1833 E16.55	Agricultural field close to BOKU, Vienna

H-high organic matter, L-low organic matter

Based on the %OC values: soil sample 1424 contains a high organic matter content, whereas soils HOC and LOC, 1425, and 1426 have a slightly lower organic content. On the other hand, among the sediments, 1419 and 1421 are rich in organic matter, while 1423 and 1427 exhibit lower organic content. From here, the conclusion is that a darker color in soils and sediments doesn’t necessarily signify a higher organic matter content.

3.2 Pesticides

I have selected six pesticides with polarities and Log*P* values ranging between 3 and 5. However, initially used tetramethrin (Log*P* = 4.6) was later excluded due to its rapid soil degradation (DT_{50} = 3). Moreover, I prepared one common solution for the investigation. All of them were of Pestanal grade. Except for fipronil, purchased from Celaflor GmbH (Bremen, Germany), all other chemicals were acquired from Sigma Aldrich (Missouri, USA).

3.3 Instrument

Applying reversed-phase HPLC and UV detection mode, pesticide levels in all solutions were determined. As HPLC can separeted both polar and nonpolar chemicals, it was suitable to use it for the determination of target components. In reversed-phase mode, the mobile phase is aqueous, containing a polar organic modifier (e.g., acetonitrile or methanol), while the stationary phase is a nonpolar (hydrophobic), such as C18 or C8-bonded silica.

Although two different types of HPLC were used for determination of pesticides concentration, the Younglin YL9100 HPLC System with YL-clarity software served as the experiment's primary tool. It has an integrated UV-VIS detector, column thermostat, vacuum degasser, quaternary pump, and YL9150 autosampler (Younglin Ltd., Anyang Korea). Compounds were separated on a C18 column (and 5µm) that has dimension of 150mm x 4.6mm at 40C by Perfectsil (Mz-Analysentechnik Ltd., Germany).

Another instrument,, ChroZen UHPLC (Young in ChroMass Ltd., Anyang Korea), has been used to record UV/Vis spectra (190nm-900nm) of the target components in order to identify the most intensive wavelength. Compared to Younglin YL9100, which is suitable to record signals only at two different wavelenths, the whole spectrum can be recorded by using ChroZen UHPLC, as it is equipped with a photodiode array (PDA) detector as well, In addition to PDA, which is sometimes called a diode array detector (DAD), this instrument has also a fluorescence detector The column used here was a C18 BRISHA (150mm x 4.6mm and 5µm) manufactured by Teknokroma Analytical Ltd, Spain.

3.4 Methods for the experiment

3.4.1 Stock and common solution preparation

First, individual stock solutions of the five pesticides were prepared at a concentration of 1 mg/mL. A specific amount in milligrams (mg) of each pesticide was carefully measured and then dissolved in a solvent appropriate for its properties by using the corresponding volume. All of the target componds (PCZ, FIP, ATC, TRF, and PDM) were dissolved in acetonitrile. Common solution containing five pesticides each at 0.2 mg/mL concentration was prepared in the required volumes.

3.4.2 Method optimization

All experiments were conducted using the same HPLC column containing C18 (Perfectsil). The column temperature was maintained at 40°C, with an injection volume of 30µL, and the eluent flow rate was set at 1.0 mL/min for all samples. During the optimization of the HPLC method, these chromatographic parameters were kept constant, eluent composition and the detection wavelengths were changed. Various gradient and isocratic eluent compositions were tested, ranging from a water mixture/acetonitrile ratio of 10:90 to 30:70. However, gradients resulted in shorter retention times, but simultaneously longer chromatographic cycles due to equilibration or baseline separation of components was not achieved. The best results were obtained using an isocratic method with a mobile phase composition of water mixture/acetonitrile (20:80), as shown in the Figure 9.

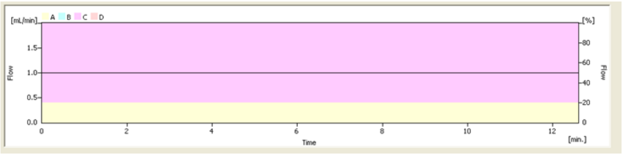

Figure 9. Mobile phase composition of isocratic method (A is HPLC grade water/MeOH (90:10), C is HPLC grade Acetonitrile)

Wavelength recordings were crucial for achieving the highest signal intensities while simultaneously eliminating possible interfering components, including the eluent. Therefore the UV spectra of individual components were recorded using ChroZen UHPLC in the range 210-220 nm at 10 µg/mL.

In this experiment, 80% of acetonitrile, with a UV cut-off at 190 nm, was used as the eluent. Optimal absorbtion has been obtained for PCZ and ATC at 210nm, FIP and TRF at 220nm, respectively. A maximum for PDM was recorded at 240nm (shown in Figure 10), but also relatively high is the absorbtion at 210 nm. Due the the close retention times for TRF and PDM, we have used the same UV wavelengths (210 and 220 nm) during whole chromatographic run to avoid uncertainities in peak areas if the retention times slightly change and ensure high signal intensities. Based on these findings, the UV detector signals were set as follows: Signal 1 is at 210 nm, and Signal 2 is at 220 nm. The total runtime for the method was 15 minutes.

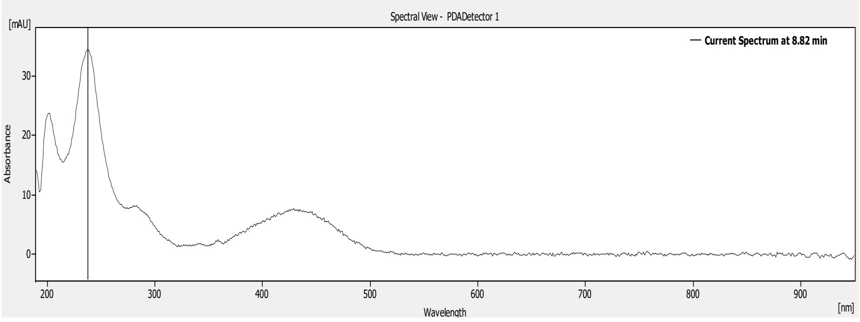

Figure 10. UV spectrum for PDM recorded at a concentration of 10 µg/mL

Peak purity was checked by calculation of peak area ratios recorde at two wavelengths (210 and 220 nm). For the individual components were as follows: ATC - 0.5423, FIP - 0.6577, TRF - 0.7276, and PDM - 0.6455.

3.4.3 Calibration

Prior to conducting measurements on the prepared solutions, it is crucial to create a calibration curve. To make calibration solutions appropriate volumes were taken from a common mixture containing all components at 200 µg/mL then further diluted with acetonitrile to yield 1mL in HPLC vials. The calibration curve was created at eleven levels: 0.02, 0.05, 0.1, 0.2, 0.5, 1, 2, 5, 10, 15, and 20 µg/ml.

A calibration graph is used to create a connection between the peak areas observed in the chromatogram and the concentrations of common solutions. Solutions, with precisely known concentrations, form the basis for the calibration curve, aligning the peak area with the known concentration. The slope, which measures peak area change per unit concentration change, is obtained from the calibration curve. This slope is crucial to the next calculations. Finally, using the slope of the calibration curve, calculate the concentration of chemicals in the samples. This is done by calculating the slope of the chromatogram by dividing the peak area by the slope of the sample. Additionally, the limits of detection (LOD) and quantification (LOQ) were calculated as follows: LOD = 3.3 x (SD intercept / slope), LOQ = 10 x (SD intercept / slope), where SD intercept = Standard error intercept * √N.

3.4.4 Sampling of pesticides in a solution

Pesticides were sampled from a solution with a concentration of 2 µg/mL spiked from a common solution of 200 µg/mL and then added to 200mL of distilled water and 10g of soil/sediment. Mixture solution was stirred for a total of 8 hours using a magnetic stirrer to achieve an equilibrium point. It was found earlier that in parallel projects 24 hours of stripping was unnecessary; therefore, a duration of 8 hours was chosen. The solution in the beaker, along with the stir bar, was stirred by a magnetic stirrer at a speed of 500-700 rpm, depending on the size of the stir bar. Beaker glasses were covered with Parafilm M by Bemis and did not use heating to prevent evaporation during stirring as illustrated in Figure 11.

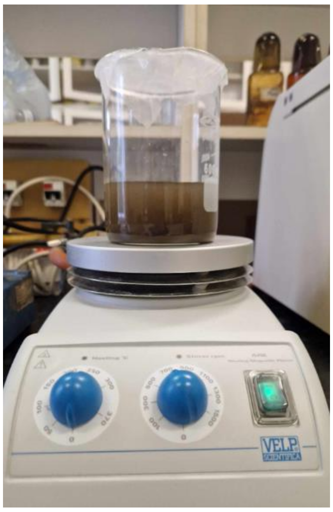

Figure 11. Stirring the solution with magnetic stirrer and stir bar

Two parallel samples were collected from the water phases at different time points: a reference sample was taken before adding soil/sediment after homogenization and then at 3, 6, and 8 hours. Liquid phase samples are taken after filtration using paper. From the stirred solution, 1.5 mL was pipetted out and placed into 1.5 mL Eppendorf tubes. (Shown in Figure 12) This step has been performed in duplicates. The water phase was then separated from the solid phase using Eppendorf centrifugation at 12000 rpm, 20°C for 10 minutes. Next, 1 mL of water phase was taken from each tube, filtered by using a syringe filter (0.45 µm PTFE), and transferred into HPLC vials. The remaining 0.5 mL solution in a tube was returned to the stirring solution.

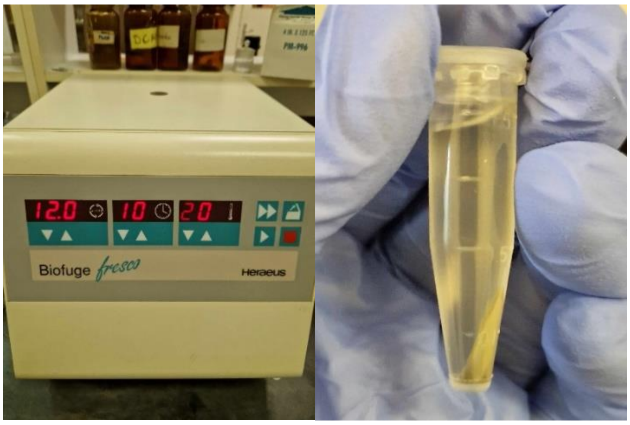

Figure 12. Eppendorf centrifuge instrument and Eppendorf tube containing separated solid phase and water phase

3.4.5 Extraction of pesticides from soil and sediment phase

After 8 hours of stirring, filtration with paper separates the solid and liquid phases (Figure 13). 10g of soil/sediment was collected from the paper. Due to the different solubilities of pesticides in water, I chose an organic solvent. These substances exhibit high solubility in organic solvents like hexane and acetone, as indicated in Table 5. Therefore, the collected 10g of soil was dried and target compounds were extracted by 15 ml of a 1:1 mixture of nonpolar hexane and polar acetone. The mixture was manually shaken for 5 minutes.

Figure 13. Separating soil/sediment phase from the water phase through a glass funnel with a paper filter and a holder

Table 5. Solubilities of the pesticides in water and organic solvents

Names	**In organic solvents at 20 °C (mg/L)**	
	Hexane	Acetone
PCZ	4700	Miscible
FIP	28	545900
ATC	-	5000000
TRF	250000	250000
PDM	49000	800000

After that, the process involved ultrasound agitation for 20 minutes, and followed by centrifugation for 10 minutes at the 3000 rpm, 4°C. 5 ml was taken from the organic phase into a 10 cm^3 test tube and evaporated to dryness in a heating module with N^2 steam, using low level of heating at 50ºC. The residue was then dissolved in 1 mL of HPLC eluents, filtered with a syringe filter (0.45 µm PTFE), and subjected to further analysis using reversed-phase HPLC. It is ideal that the residue is dissolved in the same eluents as the mobile phase in LC-UV to prevent peak broadening and tails.

3.4.6 Extraction of pesticides from the paper phase

After removing the soil/sediment, some residue remained on the papers. These papers were then cut into uniform one cm^2 pieces and placed in a centrifuge tube containing 25mL of acetonitrile, as depicted in Figure 14. Followed by ultrasound agitation for 20 minutes and centrifugation for 10 minutes at 3000rpm, 4°C. After, 5 mL of the paper extract was transferred to a test tube and evaporated to dryness using the same method outlined in the section 3.4.5

Figure 14. Paper phase analysis in 50mL centrifuge tube and centrifugation instrument

A volume of 5 mL of the organic phase was transferred into a test tube with a 10 cm^3.
The sample was then evaporated until dryness using a heating module with N2 steam at a low speed, maintaining a temperature of 50ºC.
The residue was dissolved in 1 mL of HPLC eluents, specifically a mixture of water and aceto nitrile in a ratio of 20:80. Then the solution was filtered using a syringe filter. (0.45 µm PTFE)

3.4.7 Measurement and Calculation

In summary, the experiment involved the analysis of five soils and five sediments for pesticides. The combined results from the filtered and primary samples were used to calculate the total area and determine the pesticide concentrations. In total, 102 samples were evaluated using HPLC, comprising 82 water phase samples and 20 samples for both the soil and paper phases.

Determining adsorption percentage, K_{d} and K_{oc} adhered to the methodology recommended by OECD guideline 106 for chemical testing (OECD/OCDE. Test No. 106: Adsorption , 2000). It's worth noting that OECD guideline 121, even though it is far more reliable than QSAR, could not entirely replace the 106' batch equilibrium method. (OECD/OCDE. Test No. 121: Estimation of the Adsorption Coefficient (Koc), 2001) For detailed equations, please refer to the literature review (Section 2.3.2.).

4 Result and Discussion

4.1 Optimization of the HPLC method

A variety of gradient and isocratic eluent compositions were tested, including water mixture/acetonitrile ratios ranging from 10:90 to 30:70. When gradients were used, however, retention times got shorter, and chromatographic cycles became longer because of waiting for equilibration or failing to separate components at the baseline level. The most optimal outcomes were achieved using an isocratic approach, utilizing a mobile phase consisting of a mixture of water and acetonitrile in a ratio of 20:80.

Retention time, RT determines individual components and shows their detected order in the chromatogram. Separate pesticide samples with a concentration of 10 µg/mL each were measured with HPLC to deliver the peak order. HPLC method for this experiment took place for 15 minutes in each sample with an isocratic method water mixture/acetonitrile (80:20). Pesticides were detected at the following retention times: 2.6, 2.9, 3.3, 5.4, 5.7 min for PCZ, ATC, FIP, TRF and PDM, respectively. Chromatography is shown in Figure 15

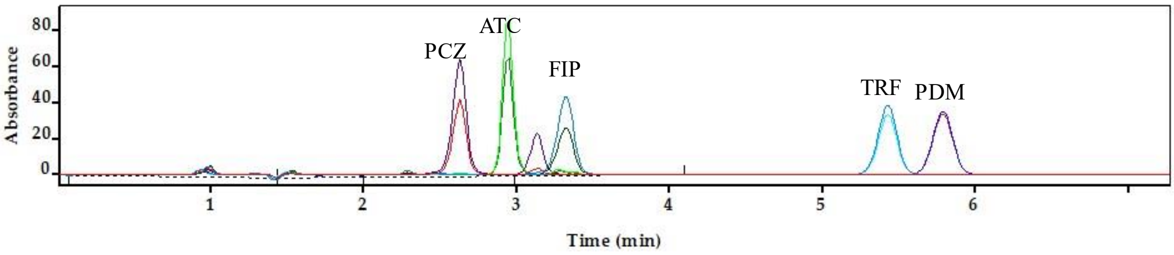

Figure 15 Retention time of each pesticide at a concentration of 10 µg/mL

As for calibration (Figure 16), each pesticide exhibits a specific slope, indicating the expected increase in detected response for a 1 µg/mL concentration increase. The number of samples with a detectable area was 11 out of 11 for ATC, 9 out of 11 for Fip, and 8 out of 11 for TRF and PDM, with no peaks at 0.02, 0.05, or 0.1 g/mL.

TRF has the highest slope of them all at 0.187, ahead of FIP (0.0767), PDM (0.0427), and ATC (0.0271). These values suggest varying sensitivities to concentration changes. While R-squared values indicate good fit, they are relatively high for all pesticides, indicating generally strong relationships. The LOD and LOQ values, representing the lowest reliably detectable and quantifiable concentrations, show minor differences between the pesticides. PRC was disqualified from further consideration because its retention time occurred too early.

Table 6 shows the slopes, linear regression values (R^2), corresponding limits of detection, and quantifications for each pesticide. It should be noted that the unit of measurement in Figure 16 is 1 part per milliom (ppm), which is equal to 1 microgram per milliliter (µg/mL), and this value is also equal to 1000 parts per billion (ppb).

Table 6. Slope, linear regression values, LOD and LOQ

Pesticides	**Slope (m)**	**R^2**	**LOD µg/mL**	**LOQ µg/mL**
FIP	0.0767	0.9894	8.0763	24.473
ATC	0.0271	0.9316	21.908	66.39
TRF	0.187	0.9875	8.808	24.448
PDM	0.0427	0.9873	8.58	26.002

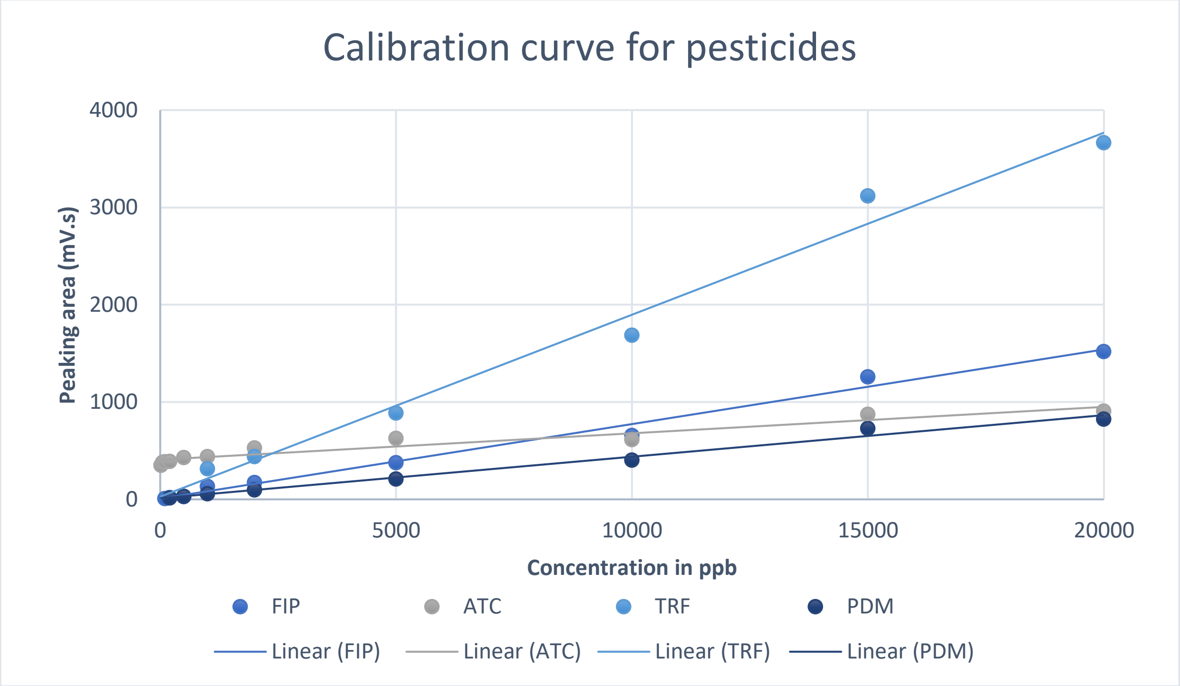

Figure 16. Calibration curves of all 4 pesticides

4.2 Concentration as a function of equilibration time

The graph in Figure 17 shows how the amount of pesticides in the water phase changed over time in Vienna soil HOC samples. The concentration were measured in parts per billion (ppb). The term "0 hour" refers to the initial homogenized sample when no soil or sediment was added. The abbreviation "LP" represents the liquid phase that occurs following the process of filtering paper. At the initial measurement (0 h), a slightly higher level was observed for all pesticides, indicating the actual quantity applied. The following calculations were adjusted based on the initial level. All pesticides demonstrated a decline in concentration within the water phase. The decreasing trend in concentration indicates that the adsorption process for soil and sediment particles is in progress.

TRF not only showed a smaller percentage in here but also did not exhibit any presence in the liquid phase after filtration, indicating it was bound to the solid phase. However, in other samples TRF showed in water phase after filtration. The PDM was more found in sediment compared to soil in other samples, especially after the filtration. All pesticides exhibited a decrease in concentration from 6 hours to the LP, except for ATC, which showed an observed increase in concentration from 8 hours to the LP.

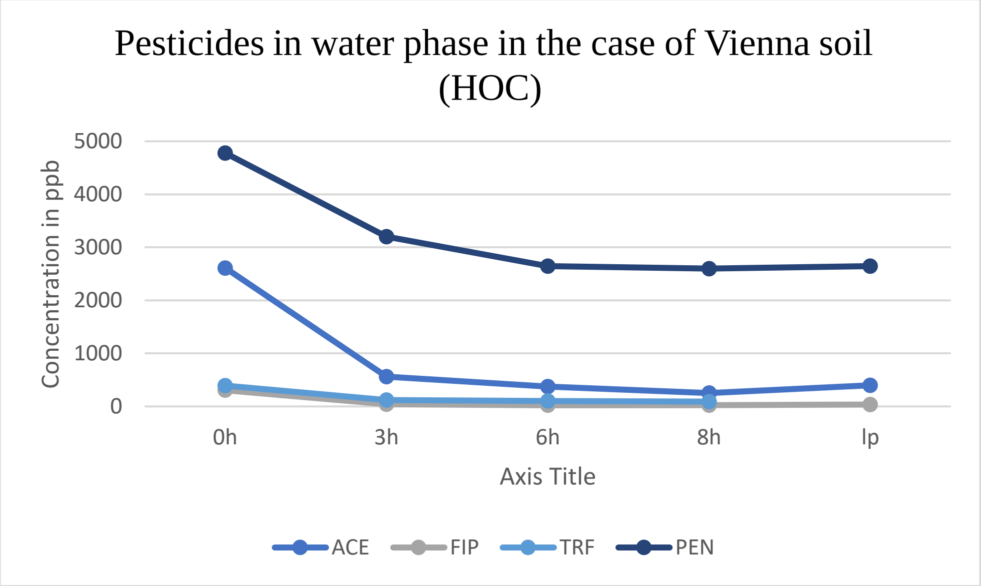

Figure 17.Concentration of pesticides in the water phase as a function of equilibration time in Vienna soil (HOC)

4.3 Adsorption % as a function of equilibration time

The figures from 18 to 21 display the adsorption percentage on the solid phase versus equilibration time for pesticides in all soils and sediments. These figures include the adsorption percentage at the specified times (A(t_i)). A (t_i)% illustrates the overall trend of adsorption during stirring. The quantities absorbed into the solid phase at specific times (t_i = 3/6/8 hours) were determined from the concentrations in the water phase, assuming that the amount that disappeared from the water was bound to the solid. A(t_i)% was calculated by dividing the amount sorbed to the solid phase at the specified time (t_i = 3/6/8 hours) by the initial amount found in the reference sample (m_o), presented as a percentage. A_{eq} % shows us the adsorption to solid phase at the equilibrium. A_{eq} % values are given in Table 7. (see Section 2.3.2).

The presence of negative values in the data might potentially be explained by evaporation, which may lead to increased concentrations in the water phase. As a result, this might potentially give rise to the perception of adverse adsorption values. Potential decomposition may also result in a negavite value. (Experimental error). As an additional precaution, we also analyzed the paper phase used for filtration due to suspicions that some elements may have been bound to it. (see Section 4.5)

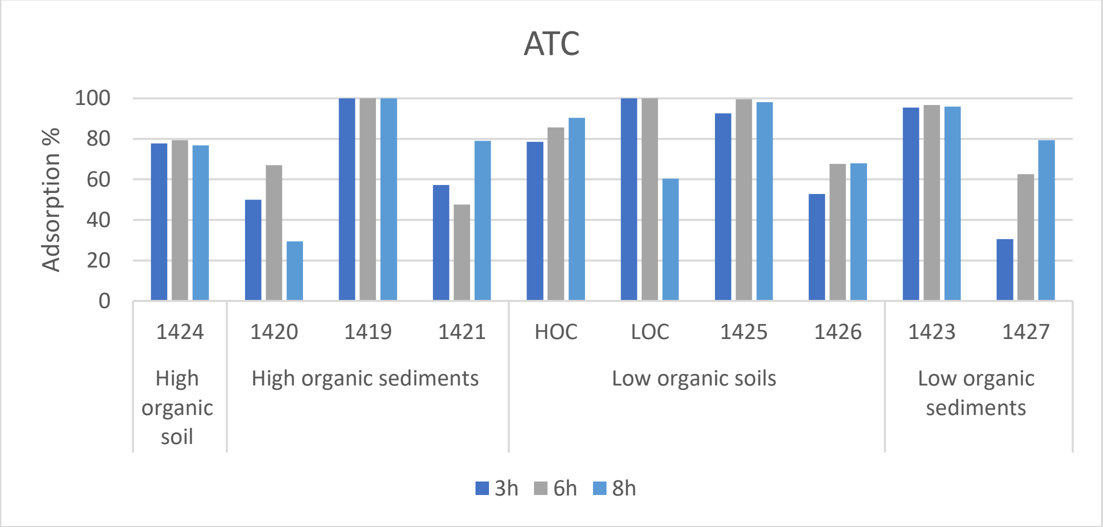

Figure 18. Equilibration Plot of Adsorption % for ATC in all soils and sediments

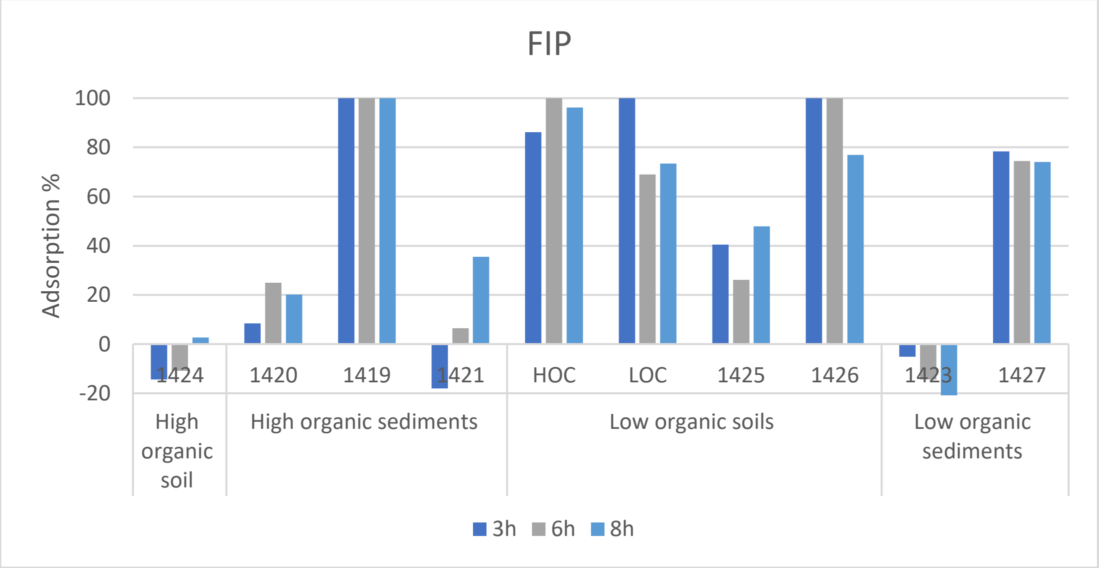

Figure 19. Equilibration Plot of Adsorption % for FIP in all soils and sediments

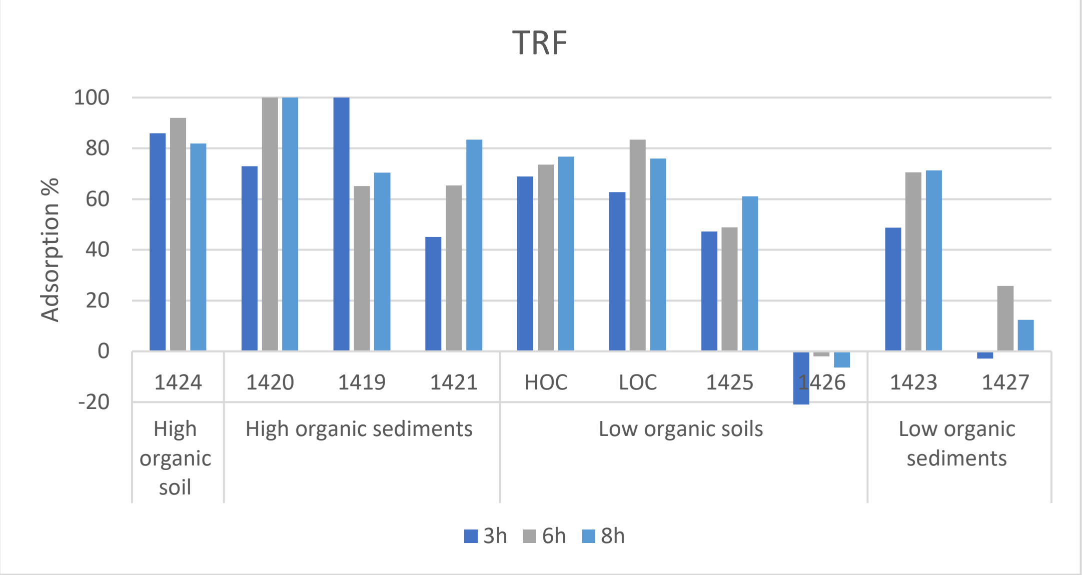

Figure 20. Equilibration Plot of Adsorption % for TRF in all soils and sediments

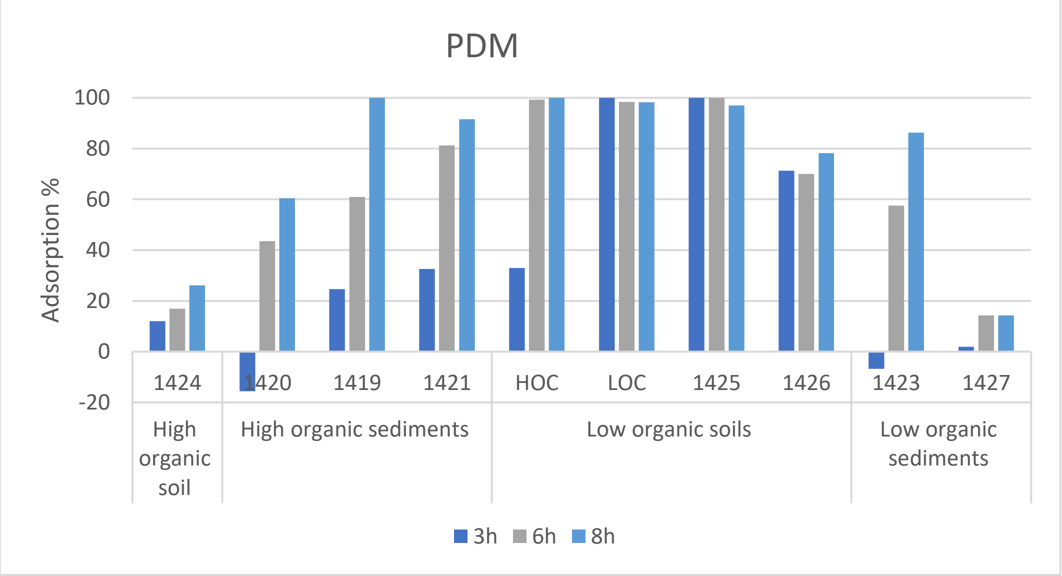

Figure 21. Equilibration Plot of Adsorption % for PDM in all soils and sediments

Almost all of the soils show an increasing trend from 3 to 6 hours. However, at 6 hours to 8 hours, the correlation varies depending on the pesticide. ATC is mostly found in the water phase rather than the solid phase. ATC may have decomposed, as shown in Figure 18, especially at 6 h, when it increases in most samples.

FIP is found equally in the water and solid phases. TRF is found more in higher organic matter soil/sediment. Furthermore, PDM was only detected in the water phases of the soil samples but was found in both the water and solid phases of the sediments. This suggests a possible relationship between particle size and mineral content (see section 2.3.3)

Despite being nonpolar components that are typically prone to binding with soil, we did not find the entire spiked amount in the solid or paper phases for some cases of TRF, PDM, and ATC. During stirring, these pesticides were not fully bound to soil particles; instead, they may have been precipitated or decomposed. Particularly, a visible distinction emerges when comparing soil and sediment samples, with the water phase of sediments exhibiting higher percentages of pesticide presence. Based on this information, it can be inferred that soils and sediments with higher organic matter content may exhibit higher adsorption capacities. However, we need to examine the distribution coefficients for adsorption (K_d) and the organic carbon coefficient (K_{oc}), as calculated in Section 4.6 to check them. The order of A_{eq}% in Table 6 from highest to lowest for the pesticides was as follows: TRF (73.91%) in 1424, High Organic Soil > PDM (21.02%) in 1427, Low Organic Sediments > FIP (10.95%) in 1427, Low Organic Sediments > ATC (0.63%) in 1421, High Organic Sediments

Table 7. Adsorption % at equilibrium for each pesticides in all soils and sediments

	High Organic soil	**Low organic soils**				**High organic sediments**			**Low organic sediments**	
	1424	HOC	1425	LOC	1426	1419	1420	1421	1423	1427
% OC	17.87	4.1	3.04	3	2.94	15.31	16.05	14.83	5.75	2.37
ACT	0.035	3.888	-	0.035	-	0.069	0.638	-	-	-
FIP	-	3.024	10.858	1.043	5.595	1.650	3.162	4.011	10.954	2.139
TRF	73.918	0.163	-	2.856	-	0.925	8.998	11.877	-	-
PDM	-	-	-	-	-	1.141	-	0.762	21.029	1.647

4.4 Standard deviation

Standard deviation values for two parallel samples determined from water phase, as well as the ranges and its percentages (SD %) were calculated. These values are given in Table 8 for all pesticides.

The percentage of paper phase found in samples is less than the RSD% (SD range %) percentage, therefore it is negligible in most of the cases

Table 8. Standard deviation between the parallel water samples of the pesticides

Pesticides	SD range %	Average SD %
ATC	0.04 - 4.45	0.75
FIP	0.05 - 4.01	0.51
TRF	0.03 - 4.35	0.73
PDM	0.07 - 2.66	0.65

4.5 Distribution of pesticides throughout the phases

The concentrations of pesticides were determined based on peak areas observed across three phases: water (wp), soil/sediment (sp), and paper (pp). Tables 9 and 10 display the average percentages related to these phases, classified based on the organic composition of soils and sediments (high or low). The calculations employed amounts (expressed in µg) obtained from water phases following paper filtration, as well as samples taken from solid and paper phases. To show how the pesticides were partitioned between phases, the average concentrations (in µg) at equilibrium for each pesticide in soils and sediments were divided by the amount that was in the water phase before the soil or sediment was added. This was then shown as a percentage. The average percentages were calculated using the following equation:

$$average\ \% = \frac{\sum_n wp; sp; pp}{n} \div m_o$$

m_o – initial amount of pesticides before adding soil and sediments

n – number of samples

Exceptionally, in the case of high organic soil (1424 in Table 14), being the sole sample with high organic matter, the concentrations of individual pesticides are presented as percentages. The data provided in Tables 8 and 9 shows significant differences in pesticide concentrations among the water, soil, and sediment phases.

Table 9. Average percentage of pesticides in water, solid, paper phases in soils

	High organic soil 1424			**Low organic soils (HOC, LOC, 1425, 1426)**		
	WP	SP (A_{eq} %)	PP	WP	SP (A_{eq} %)	PP
ACT	26.79%	0.03%	0.63%	17.55%	0.98%	2.48%

FIP	84.88%	-	5.74%	32.93%	5.13%	0.53%
TRF	[a]55.53%	[a]73.92%	-	35.61%	36.97%	1.73%
PDM	-	-	0.19%	10.82%	-	0.12%

[a] *peak purity checked and found interacting components.*

Table 10. Average percentage of pesticides in water, solid, paper phases in sediments

	High organic sediments (1419, 1421,1420)			**Low organic sediments (1423,1427)**		
	WP	SP (A_{eq} %)	PP	WP	SP (A_{eq} %)	PP
ACT	82.45%	0.24%	1.41%	24.27%	-	2.45%
FIP	86.88%	2.94%	2.71%	81.69%	6.55%	0.89%
TRF	14.23%	7.27%	-	32.27%	-	-
PDM	31.39%	0.63%	-	38.53%	11.34%	-

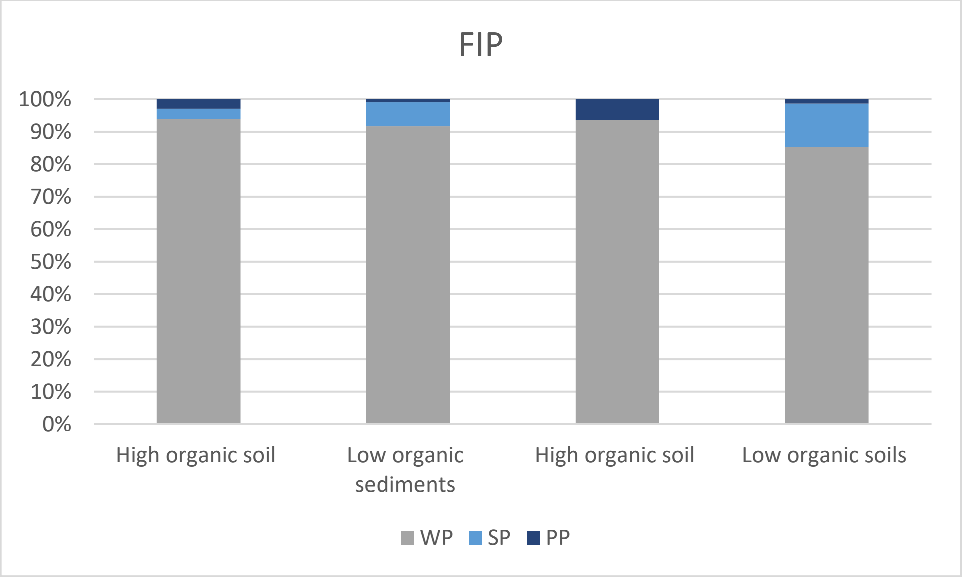

Figure 22 illustrates the distribution of FIP among the water, solid, and paper phases

As previously mentioned, the recoveries of ATC, TRF, and PDM did not reach 100 percent. ATC was mainly observed in the water phases, with a minor presence in the paper phase. Although FIP was primarily detected in the water phases, it was also found in the solid phase at a lower concentration. TRF was found in both the water and solid phases, especially in organic-rich soil, where concentrations were highest. Particularly, TRF was absent in the paper phases. Upon closer analysis of certain (1424) soil samples that had unusually high concentrations. We conducted a check of peak purity, which resulted in a finding of interacting compounds.

PDM was primarily found in sediments rather than soil, and its detection was limited to soils characterized by low organic content. Due to the difficulty in recovering most of it, we suspect this could be attributed to prolonged storage, lasting 3–4 weeks in colder temperatures. To test this hypothesis, we examined a spiking mixture. The results revealed that ATC is subject to decomposition and instability, while FIP and TRF showed lower concentrations than expected. Contrary to expectations, PDM exhibited stability in organic solvent. However, this is very different from the water-phase solution of PDM.

In an effort to determine the freezing point or lowest storage temperature of the pesticides used, information from both Celaflor GmbH and Sigma Aldrich is unfortunately unavailable. Given that freezing points and storage recommendations can vary significantly depending on the manufacturer, the search for accurate data becomes crucial.

From details provided by other manufacturers, it appears that FIP is the most stable at colder temperatures, followed by PDM and TRF, with ATC being the least stable. (Frederick, (n.d)) (Safety Data Sheet: Fipronil standard (n.d)) (Pohanish, 2015) Hence, FIP demonstrates the highest recovery. (Figure 22)

Additionally, a new substance probably a metabolism appeared at 14.8 minutes, in the spiking solution creating uncertainty. Further investigation is needed to understand what it is and how it affecting the observation.

Based on these findings, it is evident that the stability of the compounds declines when exposed to lower temperatures. It is advisable to conduct measurements immediately following the completion of sample preparation.

4.6 Distribution coefficient for adsorption K_d and organic carbon-water partition coefficient K_{oc}

The K_d and K_{oc} values presented in Table 11 were calculated using the formula outlined in Section 2.2.2. For K_d, it involved the division of the quantity determined in the solid phase by the quantity in the aqueous phase at equilibrium. In Table 12, the K_{oc} values were derived by multiplying the K_d values by 100% and then dividing them by the organic carbon content (OC).

Each pesticide exhibited distinct behavior in various soil and sediment types. The order of A_{eq}%, K_d, and K_{oc} values demonstrated a positive correlation with the Log$P$ values of the studied pesticides; higher Log$P$ values corresponded to greater adsorption properties. PDM, TRF, FIP, and ACT showed the highest K_{oc} values. TRF had the highest K_d, followed by PDM, FIP, and ACT in that order.

While the trend generally aligns with the Log*P* values in Table 2, the K_d and K_{oc} values for TRF and PDM were significantly lower than the range reported in the literature. Only ACT and FIP's K_d and K_{oc} values fall within the expected range. However, it has to be pointed out that the literature data on K_d and K_{oc} values for TRF and FIP comes from different individual studies and is not used for regulatory purposes. ATC and PDM data are validated by the EU Pesticides Database and the European Food Safety Authority (EFSA).

However, ATC was not detected in some solid phases, making it challenging to calculate K_{oc} and K_d. PDM did not show a significant presence in the water phase of soils, while, on the other hand, TRF did not appear in some solid phases. Due to these factors, the calculation of most K_d and K_{oc} values of PDM and TRF was not possible.

TRF and PDM were found in small amounts in the water phase, or not at all, consistent with their low water solubility and high Lo*gP* values. FIP is likely to bind regardless of organic matter, soil, or sediment type. In contrast, ACT and TRF are more likely to bind in soils and sediments with higher organic matter content. On the other hand, PDM is more commonly found in sediment and was not observed in the water phase of soils. These results provide insights into how these pesticides interact with different types of soil and sediment.

Table 11.Values of K_d (mL/g) for pesticides in 10 soils/sediments

	High Organic soil	**Low organic soils**				**High organic sediments**			**Low organic sediments**	
	1424	HOC	1425	LOC	1426	1419	1420	1421	1423	1427
% OC	17.87	4.1	3.04	3	2.94	15.31	16.05	14.83	5.75	2.37
ACT	0.038	5.764	-	0.025	-	0.035	0.239	-	-	-
FIP	-	4.576	6.307	0.419	4.143	0.352	0.679	1.352	3.089	0.513
TRF	28.223	-	-	-	-	3.161	9.317	15.740	-	-
PDM	-	-	-	-	-	1.641	-	0.203	6.185	7.013

Table 12. Values of K_{oc} (mL/g) for pesticides in 10 soils/sediments

	High Organic soil	**Low organic soils**				**High organic sediments**			**Low organic sediments**	
	1424	HOC	1425	LOC	1426	1419	1420	1421	1423	1427
% OC	17.87	4.1	3.04	3	2.94	15.31	16.05	14.83	5.75	2.37
ACT	0.211	140.582	-	0.849	-	0.230	1.482	-	-	-
FIP	-	111.616	207.483	13.967	140.903	2.298	4.230	9.119	53.722	21.635
TRF	157.933	-	-	-	-	20.649	58.049	106.138	-	-
PDM	-	-	-	-	-	10.719	-	1.371	107.558	295.898

Even though PDM exhibits stability at colder temperatures and remained stable in organic solvent after measuring the spiking mix, it did not appear in most water phases, possibly due to its low WS (DT_{50} = 16 days) and WP (DT_{50} = 4 days). Furthermore, ATC's SD (DT_{50}) is quite low, being only 14 days. Additionally, TRF's WS (DT_{50} = 5.5 days) is also low. These factors probably affected the results.

5. Conclusions and Recommendations

In conclusion, the study highlighted distinct behaviors of pesticides, with TRF and ATC being more prevalent in soils with higher organic matter content, while PDM exhibited a likely association with sediment. Despite expectations, TRF, PDM, and ATC were not fully recovered during stirring, possibly undergoing precipitation or decomposition. Paper phase concentration was negligible. The solutions were stirred for a duration of 8 hours, when equilibrium was achieved for each pesticide. Interacting compounds were identified during peak purity checks.

ATC demonstrated the least stability at cooler temperatures, followed by PDM, TRF, and FIP. Consequently, FIP exhibited the highest recovery, emphasizing the impact of lower temperatures on compound stability. Further analysis of the distribution coefficients (K_d and K_{oc}) provides valuable insights into the adsorption capacities of the pesticides. The positive correlation between A_{eq}%, K_d, and K_{oc} values and Log$P$ values indicated better adsorption for pesticides with higher Log$P$ values. Although the trend generally aligned with Log$P$ values, TRF and PDM's K_d and K_{oc} values were considerably lower than the literature ranges. Only ACT and FIP's values fell within acceptable ranges. PDM's absence in the water phase was likely due to its low WS and WP (DT_{50}), and TRF's low WS (DT_{50}) and ATC's low SD (DT_{50}) which may influence the results.

In the proposal, method validation across varied soils and sediments is important, especially considering that recoveries have not been determined. In particular, instances of higher concentrations were found, encouraging more research. Furthermore, it is recommended to carry out 3 or more parallel samples.

To conduct a thorough examination, we propose evaluating both the used parafilm and the utilized beaker. These components should be washed with suitable solvents, such as acetonitrile or methanol, depending on the solubility of the test substance. The results will then be tested. In a previous parallel project, an examination of the used filter revealed no significant percentage of the substance.

It is recommended to perform the partitioning determination immediately following the preparation of the samples in order to ensure precise measurements and improve the reliability of the results.

6. Summary

This research aims to replicate a scenario in which surface water, containing pesticides, interacts with the sediment phase. Pollutants are partitioned between the various phases within the water, and soil/sediment system. The process of adsorption, which involves the movement of a solute from the aqueous phase to the solid phase, plays an important role. Additionally, adsorption is closely linked to how pesticides move within the soil phase and the potential for groundwater leaching, as well as the overall pollutant mobility within a river system. It's vital to investigate this process in connection with soil and sediment characteristics, particularly the organic matter content. Hence, this study explores the adsorption characteristics of four pesticides, each possessing different polarities (Log*P* is between 3 to 5) when introduced into a solution of water and soil or sediment. Understanding the time required for individual pesticides to reach equilibrium is one of the points in this thesis.

The solutions containing the pesticides at a concentration of 2 µg/mL in 200 mL of distilled water, were then slowly stirred with 10g of soil/sediment to reach equilibrium and analyzed with HPLC-UV. The investigation of the four pesticides was performed by using 10 soils with a solid-liquid extraction method. Three phases were analyzed: water, solid (soil and sediment), and filter paper. Depending on the test substance's water solubility and Log*P* value, we could expect a distribution of pesticides between the phases. So far, the distribution coefficient for adsorption K_d and the adsorption percentage at equilibrium have been calculated.

PDM exhibited the highest adsorption, but only in sediments. TRF and FIP consistently showed the highest adsorption in most cases, as PDM did not appear in water phases, likely due to its low water-sediment dissipation time at half is being 16 days. ATC, with its low soil degradation dissipation time at half of it's orginal amount left only 14 days, was more present in water. The observed correlations indicated that pesticides with high Log*P* values and low water solubility exhibited higher adsorption rates. However, it's important to note that these factors don't always have a strict connection but usually show a pattern.

There was no strict correlation between soil and sediment organic matter (% OC) content and the adsorption properties of the pesticides, except for PDM, TRF, and ATC, which were found more in higher organic soil/sediments. Interestingly, FIP showed no correlation between organic matter and adsorption. Particularly, there were differences between soil and sediments, with PDM mainly found in sediment and not in soil, possibly due to variations in mineral content and particle sizes.

These pesticides are not very stable desptice of cold storage conditions, emphasizing the need for immediate testing after preparation. Overall, the study highlights the complex interactions between pesticides, organic matter, and storage conditions, providing insights into their adsorption properties in different environmental matrices.

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8.Table of Tables

9.Table of Figures

10. Appendix

Appendix A Corresponding definitions of the symbols with their units for the 5 equations in section 2.3.2

Symbol	Definition	Units
$\boldsymbol{A_{t_i}}$	adsorption % at the time point (t_i)	%
$\boldsymbol{A_{eq}}$	adsorption % at equilibrium	%
$\boldsymbol{m_s^{ads}(t_i)}$	mass of the substance adsorbed on the soil (solid) at the time point (t_i)	µg
$\boldsymbol{m_s^{ads}(eq)}$	mass of the substance adsorbed on the soil (solid) at adsorption equilibrium	µg
$\boldsymbol{m_{aq}^{ads}(eq)}$	mass of the substance in the aqueous phase at adsorption equilibrium	µg
$\boldsymbol{m_{aq}(t_i)}$	mass of the substance in the aqueous phase at the time point (t_i)	µg
$\boldsymbol{m_o}$	mass of substance at the beginning of the test	µg
$\boldsymbol{m_{soil}}$	dry mass of soil	g
$\boldsymbol{K_d}$	distribution coefficient for adsorption	cm^3 g^{-1}
$\boldsymbol{K_{oc}}$	organic carbon adsorption coefficient	cm3 g^{-1}
$\%\boldsymbol{oc}$	percentage of organic carbon in the soil sample	g g^{-1}
$\boldsymbol{C_s^{ads}(eq)}$	concentration of the substance adsorbed on the soil (solid) at adsorption equilibrium	µg g^{-1}
$\boldsymbol{C_{aq}^{ads}(eq)}$	concentration of the substance in the aqueous phase at adsorption equilibrium	µg g^{-1}
$\boldsymbol{V_0}$	initial volume of the aqueous phase in contact with the soil	cm^3

9. Abbreviations

ACT: Acetochlor

DT_{50}: Dissipation time at half

EC_{50}: Concentration that induces 50% of the substance maximum effect

FIP: Fipronil

HPLC: High-Pressure Liquid Chromatography

K_d: Concentration of chemical in soil/Concentration of chemical substance in water

$\mathrm{K_f}$: Freundlich solid-water distribution coefficients

K_{oc}: Organic carbon-water partition coefficient

K_{ow}: Octanol-water partition coefficient

LC: Liquid Chromatography

LC_{50}: Concentration required to kill 50% of the test substances

LD_{50}: lethal concentration 50%

LOD: Limit of detection

Log$P$: Logarithmic value of octanol-water partition coefficient

LOQ: Limit of quantification

MC: Moisture Content

NOAEL: No Observed Adverse Effect Level

NOEC: No Observed Effect Concentration

OC: Organic Carbon

OM: Organic Matter

PCZ: Propiconazole

PDM: Pendimethalin

PP: Paper phase

SD: Soil degradation

SP: Soil phase

TRF: Trifluralin

UV: Ultraviolet

WP/LP: Water Phase, Liquid Phase

WS: Water-Sediment

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Sincerely,

Bulgansor Shinebayar

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