Impacts of (Nano)formulations on the Fate of an Insecticide in Soil and Consequences for Environmental Exposure Assessment

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Supporting Information

ABSTRACT: The development of nanopesticides has recently received an increased level of attention. However, there are very few data about the environmental fate of these new products, and it is not known whether nanoformulations can be evaluated within the current pesticide regulatory framework. Sorption and degradation parameters of the insecticide bifenthrin were measured in two soils for (i) the pure active ingredient, (ii) three nanoformulations, and (iii) a commercially available formulation. In most cases, fate parameters derived for the nanopesticides were significantly different from those derived for the pure active ingredient (factors of up to 10 for sorption and 1.8 for degradation), but discrepancies were not easy to relate to the characteristics of the nanocarriers. In some cases, differences were also observed between the commercial formulation and the pure active ingredient (factors of up to 1.4 for sorption and 1.7 for degradation). In the regulatory context, the common assumption that formulations do not influence the environmental fate of pesticide active ingredients after application seems therefore not always adequate. In the absence of direct measurement, an inverse modeling approach was successfully applied to evaluate the durability of the formulations in soil (release half-life ranged between 11 and 74 days). Predicted groundwater concentrations very much depended on the modeling approach adopted but overall suggest that the nanoformulations studied could reduce losses to groundwater.

1. INTRODUCTION

Applications of nanotechnology for agriculture have recently received a great deal of attention, with a particular interest in developing so-called “nanopesticides”. The term nanopesticide encompasses a great variety of products covering a wide range of properties and levels of development.1 Considering current market constraints (e.g., stringent regulations and low costs of current agrochemicals), the nanopesticides most likely to emerge in the near future will consist of new formulations of existing active ingredients (AIs).2,3

All pesticide AIs need to be formulated for specific applications, and many existing agrochemical formulations unintentionally contain structures in the nanometer size range.4 Developments of nanotechnology over the past decade have allowed formulation scientists to gain a better understanding and better control of structures at the nano scale and allowed an improvement in formulation design to suit a given purpose.

In most parts of the world, pesticides are subject to a very strict regulatory authorization process. The possible impact(s) of formulations on the environmental fate and the effect of AIs in pesticides have, however, been evaluated to a limited extent until now.3 For instance, it is often assumed that formulations and AIs separate rapidly upon application in the field.3 While this assumption is probably acceptable for some formulations, its validity should be verified for nanoeenabled formulations that are designed to modify the environmental fate of AIs. An adequate exposure assessment of nanopesticides is essential for assessing the new risks and new benefits associated with these new products. There are, however, very few available data about the behavior of the nanopesticides after their application in the field, and it is not known whether nanopesticides can be adequately evaluated within the current pesticide regulatory framework.6

A study was previously conducted with a polymer-based nanoformulation of atrazine, a herbicide AI characterized by very weak sorption and moderate degradation in soil. Laboratory experiments showed that the nanoformulation could slightly increase the measured sorption coefficients but the nanoformulation had no effect on atrazine degradation kinetics.7 The limited impact of the nanoformulation on fate parameters was most likely due to an almost immediate release of the atrazine from the nanocarriers. In such cases, the impact on environmental fate is expected to be minimal and assessing

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exposure using fate parameters derived for the pure AI and using current tools would likely be satisfactory. Further studies that consider other combinations of nanoformulations and AIs are urgently needed, in particular with nanoformulations exhibiting longer durability upon application to soil.

We here present investigations of the sorption and degradation behavior of bifenthrin, a synthetic pyrethroid insecticide characterized by strong sorption and relatively slow degradation.5 Bifenthrin is expected to remain largely immobile after soil application and is considered to represent a relatively low risk of contamination of groundwater. However, when bifenthrin is applied as a nanoformulation, association with mobile nanocarriers could facilitate the transport of bifenthrin along the soil profile, similar to the phenomenon described for natural colloids.9,10

The aim of this study was to evaluate (i) the extent to which different formulations may affect the fate of bifenthrin in soil and (ii) how discrepancies could be integrated into fate modeling for environmental exposure assessment. Soil sorption and degradation parameters of bifenthrin were measured for (i) the pure active ingredient, (ii) three nanoformulations (polymer-based nanocarrier systems), and (iii) a commercially available formulation. Several approaches were evaluated to characterize the nanocarriers present in the nanoformulations (e.g., size distribution and surface charge) as well as their association with the active ingredients (in terms of release rate or durability). Finally, a series of exposure modeling exercises were conducted to account for the impact of formulations on fate parameters.

2. EXPERIMENTAL SECTION

2.1. Chemicals. The bifenthrin analytical standard was purchased from Fluka (Sigma-Aldrich, 98.6% pure). Analytical grade acetonitrile, acetone, and methanol were purchased from VWR, and all aqueous solutions were prepared with ultrapure water (Millipore, Elix5-Milli-Q, Gradient). Three model polymer-based nanoformulations, NFA, NFB, and NFC, were provided by Vive Crop Protection, Inc. A commercial formulation of the insecticide bifenthrin (CF) was also used. Capture LFR is a suspension concentrate intended for soil application (purchased from Harvey’s Dixie, Kinston, NC). The composition of each formulation is presented in Table S1. Suspensions were prepared for each formulation and used within 24 h.

2.2. Characterization of the Formulations. Hydrodynamic diameters and ζ potentials were determined on the basis of dynamic light scattering and electrophoretic mobility, using laser Doppler velocimetry (ZetaSizer Nano ZS, Malvern, U.K., operating at 633 nm). Backscattered light was observed at a fixed angle of 173°, and the autocorrelation function was fitted using the cumulant method, assuming a refractive index of 1.5. The ζ potential was calculated from the electrophoretic mobility using Henry’s equation. Measurements were performed with pesticide suspensions containing 1 g/L bifenthrin and prepared either in ultrapure water or in 0.01 M CaCl₂.

2.3. Soils. Two standard top soils were sampled in December 2013 by LUFA Speyer in Germany. There was no application of any fertilizers or pesticides for at least 4 years prior to sampling. The sand (2.8% clay, 86.7% sand, and 0.66% organic carbon) was uncultivated, whereas the loam (26.2% clay, 33.2% sand, and 2.21% organic carbon) was sampled from a meadow with apple trees. Sampling, sieving at 2 mm, and analyses were performed following ISO standards and Good Laboratory Practices. The main soil properties and characterization methods are listed in Table S2.

2.4. Measurement of Sorption in Soil. Bifenthrin sorption was measured following the OECD guidelines.11 Soil suspensions prepared in 50 mL glass centrifuge tubes (2 g in 40 mL of 0.01 M CaCl₂ in triplicate) were equilibrated overnight on a horizontal shaker (150 rpm). Bifenthrin was then added to reach a final concentration of 1.5 mg/kg of soil, with one of the formulations or with the pure bifenthrin diluted in acetone. The amount of acetone was kept below 0.1% to minimize cosolvent effects. The samples were returned to the shaker for 24 h to reach sorption pseudoequilibrium, based on preliminary experiments measuring sorption between 7 and 48 h, see the Supporting Information. The aqueous phase was then separated from the soil by centrifugation (2000g for 30 min), extracted thrice with 5 mL of hexane, concentrated to 1 mL, and analyzed by gas chromatography and mass spectrometry (GC–MS) (see section 2.7). Recoveries ranged from 116 to 150%, and data were corrected accordingly, using one correction factor specific to each formulation.

The soil phase remaining at the bottom of the centrifuge tubes (2 g) was extracted by adding 4 mL of acetone and shaking for 1 h at 150 rpm on a horizontal shaker. After settling for at least 2 h, the acetone supernatant was sampled and analyzed by high-performance liquid chromatography (HPLC) (see section 2.7). Extraction recoveries ranged from 102 to 122%. Sorption coefficients (Kₛ, L/kg) were calculated as the ratio of the concentration of bifenthrin in the soil phase to that measured in the aqueous phase.

2.5. Incubation To Measure Degradation in Soil. Soil degradation studies were performed in triplicate and according to the OECD guidelines.12 The fresh soils were adjusted to 40% (loam) and 50% (sand) of the maximal water holding capacity and preincubated at 20 ± 1 °C for 26 days in the dark. The equivalent of 300 g of dry soil was placed in glass jars, and bifenthrin was spiked, either with one of the formulations diluted in ultrapure water (10 g/L) or with pure bifenthrin dissolved in acetone (100 g/L), to reach a concentration of 10 mg/kg of soil. A measurement of dehydrogenase activity previously described in ref 7 (described in the Supporting Information) indicated that there was no significant influence of the addition of acetone on soil microbial activity. Upon being spiked, each soil sample was mixed with a spatula for at least 5 min to maximize the homogeneous distribution of bifenthrin. The jars were then covered with aluminum foil and incubated in the dark at 20 ± 1 °C. The soil water content was adjusted once a week by mass. At suitable time intervals, 10 g of soil was sampled from each jar and frozen at −25 °C until extraction and analysis were performed. Soil samples were extracted with acetone (ratio of 1:1) following the method described in section 2.4. Overall, 15 soil samples were taken over a period of ~320 days. The soil microbial activity was controlled regularly on samples randomly selected from the incubated jars.

Degradation curves were fitted with three kinetic models, a single first-order model, a sequential first-order model (Hockey-Stick), and a biphasic first-order model (Gustafson and Holden), following FOCUS recommendations13 and using CAKE 3.1 (Tessella Ltd.) to derive DT₅₀ values (time required for half the initial amount of bifenthrin to be degraded).

2.6. Determination of the Formulation Release Rates. A number of approaches were tested to determine the rate of
release of bifenthrin from the nanocarriers when present in soil or in aqueous solution. A useful method should allow the rapid separation of that portion of bifenthrin that is released from the portion that is still associated with the nanocarriers and should allow measurements over time. None of the approaches we experimentally tested (i.e., centrifugation, ultracentrifugation, dialysis, passive sampling, filtration, and headspace analysis, described in the Supporting Information) appeared to be suitable, mainly because of the physicochemical characteristics of bifenthrin (i.e., very high sorption and relatively low volatility).

An indirect approach previously proposed by Ford et al.\textsuperscript{14} was thus used instead. Assuming that only that portion of bifenthrin released is available for degradation, a slower degradation of formulated bifenthrin can be ascribed to the slow release of the analyte. Assuming that both the release and degradation processes follow first-order kinetics,\textsuperscript{12} the concentrations of formulated, released, and degraded bifenthrin can be described by a sequential first-order model. The degradation rate was fixed to that derived experimentally for pure bifenthrin (see section 2.5), and the release rate was derived by fitting degradation curves of the formulated bifenthrin (details about the equations are provided in the Supporting Information and Figure S1). $R_{50}$ values were then calculated from the release rates, as the time required for half the bifenthrin to be released from the nanocarriers.

2.7. Extraction and Analytical Methods. Aqueous samples were extracted thrice with hexane (8:1); extracts were concentrated to 1 mL and analyzed via GC–MS (Agilent Technologies). The column (HP-5MS UI, 30 m × 0.25 mm × 0.25 μm) temperature was held for 1 min at 55 °C, before being increased to 300 °C (15 °C/min). The flow velocity of helium was 1 mL/min, and the inlet temperature was 300 °C. Bifenthrin was quantified in selected ion monitoring mode at $m/z$ 181 (ions at $m/z$ 165 and 166 were used as qualifier ions) and based on an external calibration. Seven standards (2.5–50 μg/L) were measured at the beginning of every run, and two standards were again measured after every three samples to account for the instability of the signal. On the basis of the signal-to-noise ratio, the limit of quantification was 1.80 μg/L (corresponding to a concentration of 0.05 μg/L in the original aqueous samples).

Soil samples were extracted with acetonitrile (ratios of 1:2 and 1:1 for the sorption and degradation tests, respectively), and extracts were analyzed by a HPLC system equipped with a ZORBAX Eclipse XDB-C18 column (4.6 mm × 150 mm, 5 μm pore size, Agilent Technologies). Using HPLC allowed the direct measurement of the extracts, thus avoiding multiple sample preparation steps. The column thermostat was set to 30 °C; the flow velocity was 1 mL/min, and the mobile phase consisted of a mixture of acetonitrile and water (starting with 20% acetonitrile and increasing to 100% acetonitrile within a 22 min run). Bifenthrin had a retention time of ~15 min and was quantified at 230 nm, based on calibration curves made of seven standards (0.5–12.5 mg/L). The limit of quantification was 0.36 mg/L (based on the signal-to-noise ratio).

All statistical analysis were performed with Prism version 6.07 ($\alpha = 0.05$).

2.8. Predicted Environmental Concentrations. If a relatively persistent pesticide is repetitively applied, concentrations in soil may build up and reach a plateau (PEC$_{\text{soil plateau}}$ mg/kg). PEC$_{\text{soil plateau}}$ values were calculated for the pure and formulated bifenthrin, based on measured degradation rates, and following recommendations by FOCUS.\textsuperscript{15} Details about input parameters and assumptions made for the calculations are provided in the Supporting Information.

Simulations were also performed with the pesticide leaching model FOCUS PEARL 4.4.4\textsuperscript{16} over 20 years and for the nine standard soil/weather scenarios collectively representing agricultural conditions in the EU.\textsuperscript{17} The average annual concentration of bifenthrin in leachate at a depth of 1 m was calculated for each year and the 80th percentile recorded (PEC$_{\text{GW}}$).

The first aim of the modeling exercise (direct approach) was to estimate the extent to which discrepancies in the fate parameters measured for different formulations can impact PEC$_{\text{GW}}$. Two crops likely to receive bifenthrin were simulated, winter cereals and apples, with yearly application rates of 0.01 and 0.05 kg/ha.\textsuperscript{18} For each formulation, the lowest $K_{f}$ and longest DT$_{50}$ measured in the laboratory were combined to estimate the greatest impact that formulations may have on exposure assessment outcomes. Values were compared to those calculated for the pure AI. All parameters used are presented in the Supporting Information.

With a goal of accounting for the durability of the formulations after their applications, and simulating the slow release of bifenthrin from the nanocarriers, another set of simulations were run (slow release approach). Nanocarriers were conceptualized as parent compounds that are “transformed” into bifenthrin according to a first-order rate reaction [the “transformation” can be used to simulate the release process, based on the rate derived from our parallel degradation experiments (section 2.6)]. The model PEARL simulates the fate of solutes and is based on equilibrium distribution coefficients that are not adequate to simulate the fate of nanoparticles in the environment.\textsuperscript{19}

On the basis of filtration theory, particles with a density close to 1 are most mobile in a porous medium when their diameter is close to 1 μm. Larger particles are retarded by sedimentation, pore blocking, and interception, while smaller particles, e.g., ~7 nm as measured in ref 20 are retarded because of attachment by Brownian motion.\textsuperscript{21} In unsaturated media, particle attachment is strongly influenced by the soil solution ionic strength and pH value (among other factors), and detachment (i.e., remobilization) is also strongly influenced by the transient flow field, changes in water content, and preferential flow.\textsuperscript{22} Therefore, particles might be fully retarded or mobile, depending on the actual soil conditions. In this modeling exercise, we thus considered two extreme scenarios for simulating particle retention. As a first approximation, mostly immobile nanocarriers were simulated by setting very high sorption coefficients, while very mobile nanocarriers were considered by setting very low sorption coefficients. The “metabolite” (i.e., bifenthrin released from the nanocarriers) was given the parameters measured for the pure AI. All parameters used as inputs are presented in the Supporting Information.

3. RESULTS AND DISCUSSION

3.1. Characterization of Formulations. The hydrodynamic diameters and $\zeta$ potentials measured in different background solutions (Table 1) indicate that the three polymer-based nanoformulations exhibit different colloidal behavior. Hydrodynamic diameters measured in water increased in the following order: NFA < NFB ~ NFC < CF. The relatively large particles detected in CF probably consist of precipitates of bifenthrin with/or other formulation
3.2. Effect of the Formulations on the Sorption of Bifenthrin to Soil. Soil sorption coefficients measured in 0.01 M CaCl₂ ranged from 281 L/kg (NFC in the sand) to 7395 L/kg (NFB in the loam) (Figure 1 and Table S3). Sorption to the loam was systematically stronger than that to the sand, most likely because of the higher organic carbon content of the loam. K_d values were consistent with the literature, although values for the three nanoformulations in the sand tended to be lower than those previously reported (K_d values in the EU registration dossier range of 992–5429 L/kg). The span of K_d values was much smaller in the loam than in the sand (i.e., factors of approximately 2 and 10, respectively).

There were no clear trends regarding the effect that formulations had on the sorption of bifenthrin. In the loam, sorption increased in the following order: NFC < AI < CF < NFB. In the sand, sorption increased in the following order: NFC < NFB < NFC < CF < AI. A two-way analysis of variance (ANOVA) indicated a significant effect of formulation on sorption, but not all discrepancies were significant (Sidak’s multiple-comparison test). For instance, in both soils, K_d values

Overall, changes in size and ζ potential in the presence of Ca²⁺ indicate that the three nanoformulations were stabilized by different strategies: mainly steric (NFA), electrostatic (NFC), or both electrostatic and steric (NFB). Following the fate of carbon-based nanocarriers in soil is currently not possible because of the lack of analytical tools for detecting and characterizing the nanocarriers under conditions of environmental relevance. This study focuses on the fate of the AI bifenthrin, which is undoubtedly the most toxic ingredient of the pesticide formulations studied. In the next section, we will see whether differences in stabilization strategies can be related to differences in the fate of bifenthrin.

Figure 1. Measured sorption coefficients (K_d, L/kg) for bifenthrin, either spiked in nanoformulations (NFA–NFC), a commercial formulation (CF), or pure bifenthrin (AI). Error bars are the standard deviation of triplicate measurements. Letters refer to statistical comparisons among formulations for a given type of soil (Sidak’s multiple-comparison test).
Table 2. Degradation Half-Lives (DT₅₀ days) and Release Half-Lives (Rₛₒ₅ days) of Bifenthrin Applied in Two Soils as Nanoformulations (NFA–NFC), a Commercial Formulation (CF), or the Pure Active Ingredient (AI) (± standard deviation; n = 3)

|           | DT₅₀ (days) | Rₛₒ₅ (days) |
|-----------|-------------|-------------|
|           | loam | sand | loam | sand |
| NFA       | 227.7 ± 15.5 bc | 442.0 ± 26.0 d | 58.3 ± 8.7 b | 39.2 ± 5.3 b |
| NFB       | 182.0 ± 4.2 b | 382.0 ± 14.4 c | 42.9 ± 3.5 a | 32.4 ± 3.8 b |
| NFC       | 224.3 ± 5.2 b | 318.7 ± 21.2 b | 73.7 ± 6.4 c | 11.1 ± 3.3 a |
| CF        | 215.7 ± 4.6 bc | 370.0 ± 23.0 c | 59.0 ± 6.3 b | 36.6 ± 3.2 b |
| AI        | 125.3 ± 13.3 a | 266.0 ± 5.0 a | – | – |

“Letters refer to statistical comparison among formulations for a given type of soil (Sidak’s multiple-comparison test).

for NFA were not significantly different from those for NFB and CF.

In both soils, sorption of bifenthrin applied as CF was not significantly different from that measured for the pure AI. Laboratory tests for evaluating pesticide environmental fate are typically conducted with the nonformulated AI, and our results indicate that Kᵣ values derived from such experiments are probably representative of the commercially available suspension concentrate. Conversely, there were significant differences upon comparison of the results obtained for the nanoformulations with that of the pure AI. For instance, NFC yielded significantly smaller Kᵣ values in both soils relative to that of the pure AI. NFC also yielded Kᵣ values smaller than that of the AI in the sand, but the opposite was observed in the loam (NFB loam is the only case in which a nanoformulation yielded a Kᵣ significantly greater than that of the pure AI).

Overall, the results indicate that Kᵣ values derived from experiments with pure bifenthrin may not adequately describe the sorption behavior of nanoformulated bifenthrin.

The mechanism leading to smaller Kᵣ values is unclear and could not be related to the characteristic of the nanocarriers. For instance, nanocarriers in NFC were the most likely to become associated with the soil phase upon centrifugation by aggregation and sedimentation (see section 3.1) and thus to lead to higher Kᵣ values. However, experimental Kᵣ values were the smallest for NFC.

Overall, the results of the sorption experiments showed that the influence of formulation on Kᵣ values strongly depended on (i) the type of soil and (ii) the type of formulation. While results obtained for the commercial formulation were similar to those of the pure AI, significant differences were observed for the nanoformulations relative to the pure AI. Both lower and higher Kᵣ values were measured for the nanoformulated bifenthrin, and discrepancies could not be related to the characteristics of the nanocarriers. The adequacy of the batch sorption test should also be questioned upon investigation of such complex systems. The nanocarriers (possibly loaded with the AI) are likely subject to nonequilibrium processes, which may result in unexpected behavior under more realistic conditions.

3.3. Degradation Experiments. Degradation curves of bifenthrin in the loam and in the sand are presented in Figure S2. On the basis of R² values (all ≥0.91; residual errors of <0.6 mg/kg), first-order kinetics provided acceptable fits. However, the model clearly overestimated measured concentrations remaining in the sand during the first 50 days of incubation. A first-order multicompartiment model more closely described the data than the single first-order model did (because of the larger number of parameters, though R² values were equivalent). The differences in degradation parameters derived from the two models were negligible, and the discussion below is thus based on parameters derived from fits with simple first-order kinetics.

DT₅₀ values (Table 2) were generally greater than those previously reported in the literature, e.g., EU dossier for bifenthrin: 54.2 days < DT₅₀ < 173.7 days for laboratory studies, and 5.4 days < DT₅₀ < 267.0 days for field studies.8 Degradation of bifenthrin in the loam was always faster than in the sand, which is consistent with the higher microbial activity measured in the loam (see Figure S3). In the loam, DT₅₀ values increased in the following order: AI < NFB < CF < NFC < NFA. In the sand, the values increased in the following order: AI < NFC < CF < NFC < NFA (Sidak’s multiple-comparison test). In both soils, the persistence of bifenthrin was prolonged when it was applied as a formulation (p ≤ 0.0025), which could be explained by (i) the possible toxicity of some of the ingredients of the formulations to soil microorganisms and/or (ii) the slow release of bifenthrin from the nanocarriers (delaying the bioavailability of bifenthrin for degradation). On the basis of dehydrogenase activity, there was no apparent difference in microbial activity between soils spiked with the formulated or pure AI (see Figure S3). The limited availability of formulated bifenthrin was thus much likely the reason for slower degradation of bifenthrin when it is applied as a formulation including the commercial formulation.

It has often been suggested that sorption is a limiting process to biodegradation, i.e., degradation tends to be slower when sorption is higher, but such a link was not observed with our data set as the level of sorption of nanoformulated bifenthrin was generally lower than that of the pure AI while its degradation was slower.

Like observations made for sorption, the effect of the formulation on degradation was more pronounced in the sand than in the loam, suggesting that greater OC content (possibly through microbial activity in the case of degradation) may mask some of the effects that formulations have on the fate of the AI. For instance, in the loam, the results for the nanoformulations were similar to those observed for the commercial formulation (CF). In the sand, however, NFA and NFC resulted in DT₅₀ values significantly greater than those observed for the CF.

Overall, the results show that formulations (in particular, NFA) can prolong the persistence of bifenthrin (which is already a fairly persistent AI).

3.4. Rate of Release of Bifenthrin from the Formulations. The direct measurement of the release of bifenthrin from the nanocarriers was not possible (section 2.6). Release rates were thus derived from parallel degradation experiments with formulated and pure bifenthrin, and fits with a sequential first-order model. The assumptions behind the approach are that (i) only that portion of bifenthrin that was
released is available for degradation, (ii) at time zero, all bifenthrin was associated with the nanocarriers, and (iii) both degradation and release processes follow first-order kinetics (details about the procedure are presented in the Supporting Information). Assumption (ii) is likely not accurate, but one can assume that at time zero, the majority of bifenthrin is associated with the carrier and the fraction that is not negligible. The method was also applied for the CF, which is not designed for slow release, illustrating that the approach may be extended to the concept of durability of a formulation after its application to soil. \( R_{soil} \) values [release half-lives (Table 2)] may also be interpreted as the time during which a formulation affects the fate of the AI.

A two-way ANOVA indicated a significant effect of the type of formulation on the kinetics of release, but there was no consistent trend among the formulations. In the loam, \( R_{soil} \) increased in the following order: NFB < NFA < CF < NFC. In the sand, \( R_{soil} \) increased in the following order: NFC < NFB < CF < NFA. There is little information about the factors driving the release of an active ingredient in a soil matrix. The literature on slow release formulations of pharmaceutical substances indicates that release rates strongly depend on the combination of loaded AI, the type of carrier, and the release medium. Our results suggest that the effect of soil type may overwhelm that of formulation.

The release of bifenthrin was much faster in the sand than in loam \((p < 0.001)\). The sorption of bifenthrin was much lower in the sand than in the loam (Figure 1), and the faster release could therefore not be explained by greater sink conditions for bifenthrin \((i.e., \) higher soil sorption would deplete bifenthrin from the soil solution and could accelerate the release of bifenthrin from the nanocarriers). The faster release rate measured in the sand could not be explained by differences in the soil pH value \((5.2 \) and \(7.2 \) for the sand and loam, respectively). Acrylate-based polymers tend to swell with an increase in pH, which tends to accelerate the release of a loaded AI. The effects that soil characteristics have on release kinetics and formulation durability certainly require further systematic study considering a larger number of soil types.

### 3.5. Predicted Environmental Concentrations

The concentrations of relatively persistent pesticides that are repetitively applied may build up in soil and reach a plateau \((\text{PEC}_{\text{soil-plateau}})\). Longer DT\(_{soil} \) values lead to greater buildup, and greater \( \text{PEC}_{\text{soil-plateau}} \) values were thus expected for the formulated bifenthrin \((\text{because it was generally more persistent than the pure AI})\). Worst-case calculations based on the DT\(_{soil} \) value measured for NFA \((\text{greatest increase in DT}_{soil} \text{ relative to that of the pure AI, i.e., 1.8-fold})\) lead to an increase in \( \text{PEC}_{\text{soil-plateau}} \) of up to 1.4-fold relative to that of the pure AI \((\text{Table S4})\).

Concentrations in groundwater were derived from simulations with the pesticide leaching model FOCUS PEARL 4.4.4. In the direct approach, \( \text{PEC}_{GW} \) values were calculated on the basis of the parameters representing worst-case combinations for leaching \((i.e., \) the lowest \( K_d \) and longest DT\(_{soil} \) measured were selected for each formulation to simulate leaching in nine scenarios). For the pure AI, two cases were simulated: \( A_{\text{I}} \) maximal \((i.e., \) lowest \( K_d \) and longest DT\(_{soil} \)) and \( A_{\text{I}} \) minimal \((i.e., \) highest \( K_d \) and shortest DT\(_{soil} \)) \((\text{all inputs are presented in Table S3})\). With the exception of \( A_{\text{I}} \) minimal most values predicted by the direct approach exceeded the European regulatory threshold of 0.1 \(\mu g/L\) \((\text{Figure S4})\). \( \text{PEC}_{GW} \) increased in the following order: \( A_{\text{I}} \) minimal \(< A_{\text{I}} \) maximal \(< CF < NFA < NFB < NFC \) suggesting that formulations may increase the rate of leaching of bifenthrin \((\text{which is consistent with the lower } K_d \text{ and higher DT}_{soil} \text{ values used as direct inputs to simulate the leaching of formulated bifenthrin})\). The increase in \( \text{PEC}_{GW} \) for the formulated bifenthrin relative to that of \( A_{\text{I}} \) maximal was greater for winter cereals than for apples and ranged from \( \sim 2 \)-fold for the CF to 90-fold for NFC \((\text{based on averages across the nine scenarios see Table S5 and Figure S4})\). The greatest increase in the rate of leaching for the formulated bifenthrin was systematically recorded for the Sevilla scenario, which combines a silt loam and a very low rate of precipitation.

With a goal of considering the slow release of bifenthrin, a slow release approach was also applied. Nanocarriers were conceptualized as parent compounds that are “transformed” into bifenthrin according to the first-order release rates determined from parallel degradation experiments \((\text{see section 3.4})\). Nanocarriers were considered either immobile \((\text{Table S6 and Figure S5})\) or highly mobile \((\text{Table S7 and Figure S6})\). As opposed to the direct approach, the slow release approach with immobile carriers led to \( \text{PEC}_{GW} \) values for formulated bifenthrin that were much smaller than that calculated for \( A_{\text{I}} \) maximal and from \( \sim 1 \) to 2 orders of magnitude below the regulatory threshold of 0.1 \(\mu g/L\). Generally, \( \text{PEC}_{GW} \) values for bifenthrin increased in the following order: NFC < NFA < NFB < CF \(< A_{\text{I}} \) maximal \((\text{suggesting this time that the use of nanoformulations may reduce the risk of groundwater contamination (Figure S5)})\). Considering mobile nanocarriers instead of immobile nanocarriers led to \( \text{PEC}_{GW} \) values for bifenthrin that were \( \sim 1 \) order of magnitude greater \((\text{Figure S6})\). All \( \text{PEC}_{GW} \) values for bifenthrin remained, however, much below the regulatory threshold and approximately 2–25 times lower than values obtained for the nonformulated \( A_{\text{I}} \) maximal.

In this case, considering the fastest and slowest release rate derived for NFC and CF had only a limited impact on the \( \text{PEC}_{GW} \) \((\text{Tables S6 and S7 and Figure S5 and S6})\). However, a sensitivity analysis has previously shown that after the sorption and degradation parameters, the release profile is the third factor influencing environmental concentrations derived from the FOCUS models. Further research will thus be needed to derive approaches for studying release rates and/or formulation durability under realistic conditions and to understand their influencing factors.

One should bear in mind that PEARL simulates the transport of solutes and that \( \text{PEC}_{GW} \) values \((\text{especially those generated for the nanocarriers})\) should be interpreted with great care. The mobility of the nanocarriers was simulated by setting very high and/or low sorption coefficients and thus illustrates only two extreme cases. The adequate fate assessment of the nanocarriers in soils would require (i) realistic fate parameters \((\text{e.g., attachment efficiency and degradation})\) and (ii) a model suitable for describing and predicting the distribution of particles along a soil profile. Such a model is not yet available. The modeling exercise presented here can thus serve as a useful base for the development of pragmatic solutions that are urgently needed for an improvement of regulatory assessment.

### 4. ENVIRONMENTAL IMPLICATIONS

Sorption and degradation experiments indicated that the impact of formulations, both non and commercial, on the fate of bifenthrin can be significant and tends to be greater in soils with low organic content. Rates of release of bifenthrin from the nanocarriers could not be directly measured in soil, and an
indirect approach based on differences in degradation kinetics was used to evaluate the durability of the formulations after their application. The rate of release of the AI from the nanocarriers is expected to play a key role and should be carefully taken into account when describing environmental fate processes.

It is essential to consider that nanoenabled products may mitigate the risk associated with the current use of pesticides. A highly conservative risk assessment strategy that would solely consist of increasing safety factors to account for uncertainty is thus not recommended. Overall, this study represents the first step toward the development of approaches for evaluating the fate of nanopesticides and other organic nanoparticles in the environment.

A full assessment of the type of nanopesticides studied here requires an evaluation of their biological effects (in terms of both toxicity and efficacy) and a better understanding of the processes driving the fate of the organic nanocarriers in soil, which represent a great analytical challenge. Currently, it is not possible to detect or quantify polymer nanocarriers in the soil matrix, because of the similarity of the elemental composition. Further efforts will thus be needed to develop suitable analytical techniques that support the design of more strategic nano-enabled delivery systems for pesticides and other bioactive substances, while ensuring a robust assessment of the new risks and benefits. There are a multitude of other organic nanoparticles that are intentionally or unintentionally released into the environment (e.g., organic nanocarriers used in the food, pharmaceutical, or cosmetic industry, fragments of polymers, and other plastic residues). Improved experimental and modeling approaches for assessing their fate in the environment and how they may interact with contaminants are urgently needed.

**ASSOCIATED CONTENT**

5 Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.est.6b02477.

Additional information about the soil properties, experimental protocols, and modeling inputs and outputs (PDF)

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Notes
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