MANUFACTURING DOUBT

HOW THE INDUSTRY DOWNPLAYS TFA'S TOXICITY

Pesticide Action Network Europe

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Executive summary

TFA is an ultra-short PFAS (*per- and polyfluoroalkyl substance*) and the final breakdown product of several PFAS, and particularly PFAS pesticides. It has become one of Europe's most widespread yet overlooked pollutants. Exceptionally persistent, highly mobile and soluble, TFA travels easily through soils, accumulates in groundwater, and contaminates our drinking water and food.

The contamination is not accidental. TFA pollution is directly linked to the continued approval of 32 PFAS pesticide active substances in the EU. These pesticides release TFA as they degrade, and their use is still rising in some Member States. Despite clear legal requirements that pesticides must not harm human health or groundwater, regulators have allowed these substances to be deliberately emitted into the environment. Meanwhile, removing TFA from water is virtually impossible without resorting to reverse osmosis, a costly, energy-intensive process that also results in significant water loss.

TFA was first flagged as a risk for groundwater in pesticide assessments as early as 1998, over 25 years ago. Despite the early warning, EU authorities continued approving PFAS pesticides, allowing TFA to accumulate in groundwater and food. Meanwhile, for decades, the chemical industry has worked to cloud the picture. Through lobbying and selective interpretation of studies, it has promoted myths that TFA is harmless or even naturally occurring. These narratives are now collapsing. Requested industry studies show that TFA's long-term impacts on human health raise serious concerns. Evidence of developmental harm, including effects on foetal development, has led to a proposal to classify TFA as presumed "toxic for reproduction", i.e. impacts fertility and healthy pregnancy development, under EU chemicals law.

The European regulators are now reassessing TFA's toxicity and reviewing safe levels of exposure. PAN Europe has obtained and analysed the studies submitted by the TFA-producing and marketing companies in Europe, forming the 'TFA task force' consortium, namely BASF, Bayer, Corteva and Syngenta, to influence the regulatory process.

Our analysis reveals how this consortium has systematically downplayed evidence of TFA's harmful effects. Contrary to industry claims that TFA is toxic only at very high doses, a growing body of data consistently demonstrates that TFA causes serious developmental effects in foetuses at lower levels, as well as other adverse outcomes, including thyroid hormone disruption, liver toxicity, effects on the immune system and abnormalities in sperm quality and quantity. These are not isolated incidents, but consistent toxicological signals observed at different levels across species and studies. Yet, the industry continues to push for unrealistically high safety thresholds, relying on selective data interpretation and insisting that the developmental toxicity potential of TFA is rabbit-specific, even though its analysis has failed to prove it and the available data reveal a different story. Moreover, the industry proposes a safety drinking water value of 294 µg/L, which is more than 100 times higher than the most protective national standards.

TFA is not a minor detail in Europe's PFAS problem. It is the most abundant PFAS in our environment, originating from multiple sources, including PFAS pesticide use. Its presence represents a toxic legacy and a clear breach of our right to safe water. This report calls on regulators not to be misled by industry's attempts to downplay TFA's toxicity and to finally uphold the law: apply the precautionary principle, stop approving PFAS pesticides, and protect people and the environment from further contamination.

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Introduction

Per- and polyfluoroalkyl substances (PFAS), so-called *'forever chemicals'*, are a group of synthetic substances deliberately designed to resist breakdown. Their carbon-fluorine bonds are among the strongest known, giving them extreme persistence and mobility. This design, which made PFAS commercially successful, is now at the root of one of the most urgent chemical crises of our time. PFAS contamination has been linked to infertility, liver damage, endocrine disruption, thyroid disease, immune suppression, and several cancers. Children and developing foetuses are especially vulnerable.

Trifluoroacetic acid (TFA, CF_3COOH), an ultra-short PFAS and a breakdown product of certain PFAS chemicals, has recently emerged as a significant yet largely overlooked pollutant. TFA is highly persistent, very mobile and now widespread in the environment. It has become ubiquitous in water systems, including in drinking water. PAN Europe's investigations have found TFA in every sample of European surface water, tap water, and even most samples of bottled mineral water. We have also detected TFA in wines, with levels rising dramatically since 2010. Moreover, it has been found in plant-based products, including baby food, and even in human blood.

Alarmingly, conventional water treatment methods are ineffective at removing TFA. Currently, the only available effective option is reverse osmosis, a costly, energy-intensive and technically challenging process. Furthermore, preventing pollution at the source is far more cost-effective than investing resources in end-of-pipe clean-up.

According to recent scientific warnings, TFA poses a serious threat to planetary boundaries, as most of the TFA released today will persist in water supplies and the environment for future generations to come.

One of the primary sources of TFA contamination is the degradation of PFAS pesticides that contain at least one fully fluorinated methyl ($–CF_3$) group. PFAS pesticides are the first source of TFA pollution in agricultural areas, according to scientific research[1]. While the Pesticide Regulation 1107/2009 requires that pesticides can only be approved if it is shown they have no harmful effects on human health and groundwater, 32 PFAS pesticide active substances are currently approved in the European Union, contributing to this contamination.

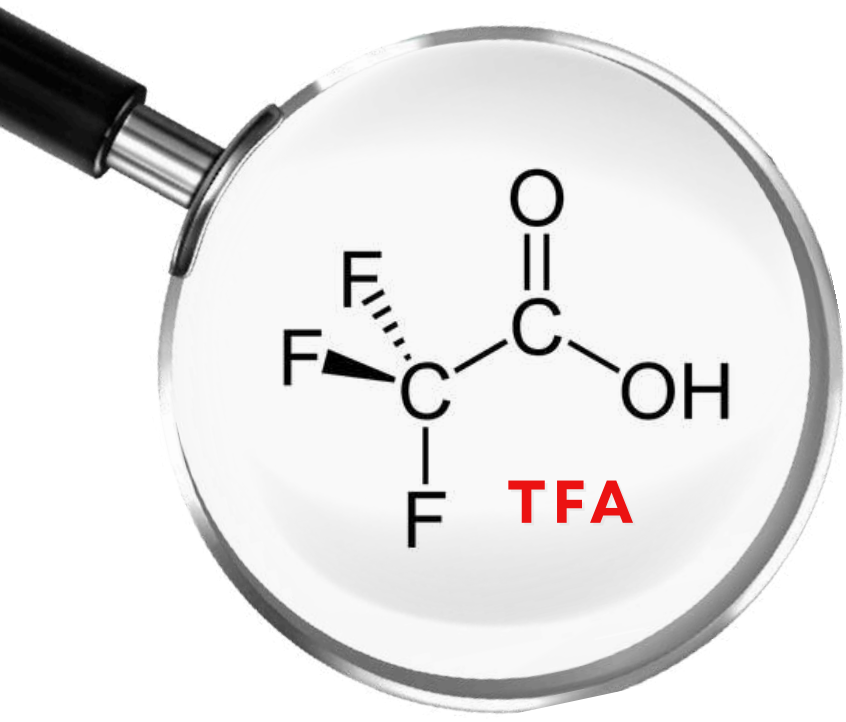
For a long time, TFA was underestimated on the assumption that it was of low toxicity. This situation changed when the European Chemicals Agency (ECHA) requested TFA producers to carry out and deliver two long-term toxicity studies on TFA (2017). Alarmingly, the studies revealed TFA's potential to induce chronic harm, especially in offspring.

[1] UBA, Trifluoroacetate (TFA): Laying the foundations for effective mitigation. Spatial analysis of the input pathways into the water cycle; Joerss H, et al. Pesticides can be a substantial source of trifluoroacetate (TFA) to water resources; FOEN, TFA in groundwater.

As a result, TFA was recently proposed for harmonised classification as “toxic for reproduction” category 1B under the Classification, Labelling and Packaging (CLP) Regulation 1272/2008. The new evidence showing that it causes reproductive toxicity makes TFA a ‘toxicologically relevant’ metabolite of PFAS pesticides, which means that by law it should not be detected above 0.1 µg/L in groundwater. This threshold is evidently already exceeded in many areas in Europe.

ECHA and the European Food Safety Authority (EFSA) are respectively being tasked to assess the hazard properties of TFA and review its toxicological reference values, i.e. levels of exposure that can be considered safe for humans. Their conclusions will have important impacts on how TFA is regulated. TFA producers, united under a ‘TFA Task Force’, have produced and submitted studies and presented their interpretation of their results to regulators, downplaying the toxic potential of TFA. By obscuring the facts from policymakers and the general public, their actions risk significantly blocking or delaying the much-needed policy measures to protect human health and the environment, now and for future generations.

PAN Europe has requested access to the industry studies submitted to the European regulators and carried our its own analysis. **Our objective was to watchdog whether the industry would attempt to cast doubt on the toxicity of the substance in order to secure weaker regulation of TFA and TFA-emitting chemicals. With this report, we aim to expose such misleading practices.**



Background

The history of TFA regulation is one of delay and denial. From 1998 to 2024, the pattern has remained unchanged: industry delays, regulators defer, and the public bears the risk.

Early warnings ignored (1998): First evidence of TFA formation in soil

The story begins in 1998, when TFA was identified for the first time as a soil metabolite of the pesticide active substance flurtamone. A study (using a lysimeter) detected consistently high average TFA concentrations over three years. At that time, the Scientific Committee on Plants[2] acknowledged a potential risk of groundwater contamination. This concern was largely sidelined: "*However, the Committee is aware of human toxicological data available from other sources suggesting possible risks to human health associated with exposure to TFAA. In conclusion, the metabolite TFAA represents a risk for contamination of groundwater but in the absence of mammalian toxicology data, the Committee was unable to evaluate the health risk*."[3]

Surprisingly, this did not lead to further action. For years afterwards, TFA remained a minor footnote in regulatory dossiers, mentioned only sporadically and never subjected to a thorough risk assessment. Attention remained limited partly because of missing data. **The industry consistently produced studies framed to downplay toxicity, rather than to fill the regulatory knowledge gaps in good faith.**

Notably, in 2007, TFA was identified by EFSA as a crop metabolite, revealing direct entry into the food chain. Nevertheless, this development did not prompt a corresponding increase in regulatory attention. In fact, most substance dossiers did not report TFA formation at all. This was not because TFA did not form, but because the degradation study relied upon (OECD 307) is ill-suited to detect it. This study fails to detect TFA depending on the analytical methods selected by industry, and because its 120-day duration does not capture the extreme persistence of TFA.

EFSA maps 39 approved pesticide active substances that form TFA (2014)

In 2014, EFSA published an analysis showing that 39 of the approved pesticide active substances at the time either break down into, or have the potential to degrade into, TFA due to their molecular structure. This concerns all PFAS pesticides that contain at least one $–CF_3$ group, known precursors of TFA.

[2] The Scientific Committee on Plants was composed of representatives of Member States and the European Commission.

[3] The average concentrations measured, 1.4 and 3.1 µg/L, are several orders of magnitude higher than the legally permitted threshold value of 0.1µg/L in groundwater, for toxicologically relevant metabolites.

Yet, the industry still avoided producing comprehensive toxicity studies or to carry out suited degradation studies. The analysis was carried out as part of EFSA's evaluation of the Maximum Residue Levels (MRLs) of saflufenacil in various crops. This included a dietary consumer exposure assessment of TFA, as a breakdown product of this PFAS active substance. Although saflufenacil has never been approved for use in Europe, its use overseas would result in residues in imported food that would exceed the default limit of 0.01 mg/kg established for non-approved pesticides. Therefore, BASF had submitted several 'import tolerances' requests for residues of saflufenacil to be allowed in EU-imported products.

A safety assessment built on gaps and guesses (2014)

The long-standing assumption that TFA posed low toxicity was **based on a highly limited dataset submitted by BASF and Bayer, which excluded the standard long-term toxicity studies** that normally form the foundation of a robust risk assessment. When EFSA established toxicological reference values in 2014, the industry had provided no animal studies on chronic toxicity, carcinogenicity, reproductive toxicity, neurotoxicity, or endocrine-disruption. Instead, the dataset consisted only of in vitro genotoxicity assays (from which EFSA concluded that TFA is not genotoxic), a poorly reported[4] developmental toxicity study in rats, and a sub-chronic toxicity study in rats. To derive an acceptable daily intake (ADI), EFSA adopted Bayer's proposed approach*: "The approach to derive the ADI of 0.05 mg/kg bw per day as proposed in the position paper submitted by Bayer CropScience (Bayer CropScience, 2013) is agreed."* [5].

In the absence of a one-year chronic toxicity study, EFSA relied on a short-term 90-day repeated-dose rat study, setting the ADI at 0.05 mg/kg. This value was extrapolated from the lowest tested dose (10 mg/kg/day) at which no liver toxicity was observed. At higher doses, however, rats exhibited clear signs of hepatic stress, including increased liver weight, hepatocellular hypertrophy (associated with peroxisome proliferation), necrosis, and elevated alanine aminotransferase levels, a key marker of liver damage or disease. Despite these limitations and the absence of critical long-term studies, EFSA applied an uncertainty factor of only 200, as recommended by Bayer and concluded that TFA posed no significant health risk. This conclusion, endorsed by the European Commission, subsequently shaped the risk assessment of all active substances that degrade into TFA.

TFA's brief recognition and quiet downgrade (2017)

Later in 2017, EFSA did briefly classify TFA as a toxicologically "relevant" metabolite of the PFAS pesticide flurtamone, because its parent compound was proposed for classification as a carcinogenic category 2.

[4] The raw data and original study report were not accessible to EFSA (according to EFSA); see EFSA 2014, p. 9. This study led EFSA to the now proven incorrect conclusion that TFA is not toxic to the unborn foetus (cf. ongoing harmonised classification of TFA as toxic for reproduction).

[5] In 2013, Bayer CropScience submitted a position paper on TFA, see EFSA 2014, p. 11.

However, this relevance classification was tied only to the parent's intrinsic properties, in line with the EU guidelines, and did not apply to other PFAS substances that are TFA emitters. When the active substance was banned, TFA's status was then quietly downgraded by EFSA and the European Commission to "irrelevant", effectively removing it from further toxicological scrutiny.

The Turning Point: ECHA demands missing data (2017)

The situation only began to shift in 2021. **Crucially, the first comprehensive toxicity studies on TFA were not commissioned voluntarily by industry, but only after ECHA compelled Solvay in 2017 to address glaring data gaps** under REACH. Until then, essential robust evidence on developmental and reproductive toxicity had simply not been produced. In 2017, ECHA, acting under the REACH Regulation 1907/2006, and following a 'compliance check' of the registration of TFA, had identified missing and/or inadequate/unfit information in the toxicity section of the substance. Thus, it formally requested that the main TFA producer conduct and submit, among others, two long-term toxicity studies on TFA: a developmental toxicity study and an extended one-generation reproductive toxicity study. It was only in response to this requirement that Solvay and Bayer commissioned the studies as co-owners. Bayer became involved because the studies would also be considered in the pesticide assessment, i.e. would impact the regulation of the PFAS pesticides marketed by the company. The results were submitted to ECHA with some delay[6].

Developmental toxicity findings in rabbits (2021)

In 2021, Bayer notified EFSA, the European Commission (DG SANTE), and Member States of preliminary results from the rabbit developmental toxicity study, as required under Article 56 of the Pesticide Regulation, which obliges companies to inform pesticide regulators of potentially harmful or unacceptable findings. The rabbit developmental toxicity study revealed both major and minor developmental abnormalities in rabbit offspring, particularly eye and skeletal abnormalities, at all doses of exposure (180-700 mg/kg/day).

Proposals for Harmonised Classification of TFA (2024)

Based on these findings, ECHA took a decisive step by inviting Member States to propose a harmonised classification for TFA as toxic to reproduction (Category 1B). While several Member States, including Sweden and Finland, initially considered preparing the classification dossier, the responsibility ultimately was taken by Germany's BAuA (Federal Institute for Occupational Safety and Health), which formally submitted the classification proposal in May 2024. At the same time, the manufacturers themselves, perhaps anticipating tighter regulation, proceeded with a self-classification of TFA as suspected of being toxic for reproduction (Category 2) under the CLP Regulation.

[6]The deadline to update the registration with the request information was 7 January 2021. Data was submitted on 8 July 2022.

Commission's recognition of TFA as 'relevant' metabolite

In light of these developments, the Commission (DG SANTE) identified TFA as a toxicologically 'relevant' metabolite of pesticides, acknowledging that it exhibits hazardous properties that are considered 'unacceptable'[7]. This designation carries significant regulatory consequences: under the the Pesticide Regulation 1107/2009 and Groundwater Directive 2006/112, a pesticide cannot be approved if its relevant metabolites exceed the threshold of 0.1 µg/L in groundwater. Alarmingly, TFA has largely exceeded this limit in numerous regions across Europe, but it has not led to an EU ban of all PFAS pesticides. The Commission (DG SANTE) requested EFSA to re-evaluate TFA's toxicological reference values, signalling the need for a more robust assessment of its health risks.

Industry submits new studies to challenge findings

In response, members of the 'TFA Task Force' marketing pesticides in the EU, namely BASF, Bayer, Corteva and Syngenta[8], suggested to the European regulators that the developmental toxicity finding *"may be rabbit specific"*[9] (and therefore not relevant to humans). The task force submitted to the European regulators a series of follow-up 'mechanistic' toxicity studies and even repeated the rabbit toxicity study starting from much lower exposure concentrations *"to fully understand the relevance of these* [rabbit] *findings"*[10]. The intention was to *"clarify species differences, to determine a mode-of-action (MoA) and to establish a No Observed Adverse Effect Level (NOAEL)"*[11] for exposure. However, not only were adverse effects found at much lower doses (i.e. 60 mg/kg/day), but the industry failed to prove that the effects are in fact rabbit-specific.

The next section will expose how the Task Force's claims rest on speculative and unsupported explanations. In fact, the evidence consistently points to TFA's potential to cause developmental toxicity across animal studies, reinforcing the case for its classification as toxic for reproduction category 1B and the establishment of precautionary toxicological reference values.

Another section also highlights how industry actors are actively advocating for drinking water limits that are substantially higher than the most protective national standards.

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[7] Cf. (11) "*(...) in accordance with the applicable Guidance documenton theassessment of the relevance of metabolites in groundwater,metabolites leaching intogroundwater whichqualify for classification under Regulation (EC) No 1272/2008 due to their reproductive toxicity, regardless of the category, are considered to be toxicologically relevant. Based on those elements, the Commission considers TFA to be a toxicologically relevant metabolite with a high potential to contaminate groundwater*." (Regulation EU 2025/910 of 30 May 2025).

[8] The Task Force also comprises Chemours, Honeywell and Solvay.

[9] 2024, TFA task force, Article 56 update.

[10] Idem.

[11] Idem.

Timeline

1998 Evidence of TFA formation in soil and risk to groundwater

2007 Evidence of TFA formation in crops

2014
- Identification of 39 approved pesticide active substances that can degrade into TFA
- Adoption of TFA toxicological reference values by EFSA

2017 Regulators identity data gaps and request long-term toxicity studies, including a developmental toxicity study

2021 Industry informs EU regulators about birth defects at all levels of TFA exposure

2024
- Germany submits a proposal to classify TFA as toxic for reproduction Category 1B
- Identification of TFA as a toxicologically relevant metabolite of pesticides

2025 EFSA proposes revised ADI and ARfD values for TFA

Overview of TFA toxicity studies and industry efforts to distort the results

This section demonstrates that the evidence of TFA's developmental toxicity is not limited to the now 'well-known' 2021 rabbit study. Other studies, including one conducted in rats, have shown similar birth defects, indicating that the effects are not species-specific. Moreover, the available studies point to additional harmful impacts of TFA, including thyroid hormone disruption, liver damage, effects on the immune system and reduced sperm quality.

Industry-Developmental toxicity studies

1.Prenatal Developmental toxicity studies

A developmental toxicity TFA study on rats (OECD 414) (see Annex nr.2) was carried out in 2010 but did not raise significant concerns from the regulators at that time. It led to a NOAEL of 150 mg/kg/day. However, important effects were observed at that dose and even lower (37.5 mg/kg/day). Namely, a significant increase in liver and kidney weights was observed in foetuses exposed to 150 mg/kg/day, while major developmental effects such as skeletal and visceral abnormalities were observed in six foetuses (including one with eye abnormalities) from this same exposure group, which were not seen in the control (non-exposed) group. At 37.5 mg/kg/day exposure group, one foetus showed a retroesophageal subclavian artery. These developmental effects were, however, considered by the industry as incidents "within Historical Control Data range" from 1992-1994, and therefore disregarded as unrelated to treatment/exposure[12]. Additional visceral or skeletal developmental effects were observed across all exposed groups (37.5, 70 and 150 mg/kg/day) but were considered minor and/or incidental and therefore they were completely disregarded and not discussed at all[13].

[12] 2010, Bayer, *"The litter incidence of malformations observed were not considered test article-related because the incidences fell within published Historical Control Data"* (page 29).
[13] 2010, Bayer, "*All other findings were considered to be infrequent, not dose-related, and consistent with normal background variation."* (page 29).

Historical Control Data (HCD) are collected from the unexposed "control" groups of previous experiments. They can be useful, particularly in long-term animal studies (e.g., carcinogenicity testing), to assess whether the control group in a new experiment is healthy and whether the overall study has been conducted properly.

If historical control data differ substantially from concurrent control data, this may indicate a broader problem with the experiment, potentially requiring the study to be repeated. Importantly, HCDs are not intended to replace the concurrent control group. Their use should be strictly limited to data derived from the same laboratory, using animals of the same species, strain, and age, and generated within the past five years.

The scientific literature consistently emphasises that concurrent control groups are the most valid, and in practice, the only valid control groups. Several studies have warned against the bias introduced by including historical controls (see Haseman, 1984; Hardisty, 1985; Cuffe, 2011).

For further discussion on the misuse of HCD, see our report: *Industry Writing Its Own Rules* (from page 30).

Serious concerns about the toxicity of TFA arose from the crucial **embryo-foetal rabbit study (OECD 414)**, which was co-sponsored by Bayer and Solvay in 2021 following the request of ECHA (see Annex nr.1). This study revealed a high incidence of major eye abnormalities (multiple folded retina and absent aqueous/vitreous humour), as well as multiple skeletal abnormalities at doses of 375 and 750 mg/kg. At the lowest dose of 180 mg/kg/day, eye malformations (absent aqueous humour, multiple fold retina, small eye) were also observed at lower incidences, showing a dose-response relationship. Foetal/pup body weight decreased significantly at 275 and 650 mg/kg/day, whereas liver weight increased, and hypertrophy was observed across all exposures. Bayer mentioned that the eye malformation incidents seen in foetuses at 180 mg/kg bw/day were within historical control data[14] but nevertheless a NOAEL for foetuses could not be established in this study as there was a dose-response relationship across exposures. Indeed, the study report clearly states that *"major foetal abnormalities were observed at all dose levels"*, and that *"the foetal NOAEL could not be established on this study."*[15]

NOAEL (No-Observed-Adverse-Effect Level) refers to the highest dose or exposure of a substance at which no harmful (adverse) effects are observed in the test population, typically in animal studies.

[14] 2021, Bayer-Solvay, page 34.
[15] 2021, Bayer-Solvay, page 34.

A follow-up rabbit study, co-sponsored by TFA Task Force members at that time (BASF, Bayer, Solvay/Rhodia and Syngenta) was conducted on its own initiative, using lower doses of exposure (see Annex nr.9). According to Bayer, the follow-up study aimed to understand the 'human relevance' of the foetal effects observed in the 2021 study, clarify species differences, determine the mode/mechanism of action and establish a NOAEL.

Human relevance is a risk assessment criterion that examines whether an adverse effect observed in animal studies applies to humans. Due to the evolutionary similarity between humans and other mammals, adverse effects observed in rodents (such as rats and mice), rabbits, dogs, and other species are generally considered relevant for humans.

However, animals are not identical to humans, and biological differences may exist. For decades, the industry has challenged evidence showing that certain chemicals are carcinogenic, often attempting to discredit results by arguing that a safe level of exposure (a threshold) exists, or effects seen in animals are not relevant to humans.

To clarify this situation, the mode of action (MoA) observed in experimental animals is compared with that in humans (often using in vitro data). This comparison helps determine whether the effects seen in animals are predictive of human risk. Unless it can be clearly demonstrated that the underlying MoA does not operate in humans, any adverse effect observed in animal studies is considered relevant for human health.

Mode of Action (MoA) refers to the biological mechanism through which a chemical substance produces an effect in a living organism. In toxicology it describes the sequence of events at the molecular, cellular, and tissue levels that ultimately lead to an observed adverse outcome, such as cancer, reproductive toxicity, or organ damage.

This new OECD 414 study included lower exposure doses (30 and 60 mg/kg bw/day), as well as a medium range (250 mg/kg/day) and high range (750 mg/kg) dose. As in the previous study, incidents of major eye malformations (folded retina and absent aqueous/vitreous humour) in offspring were observed at higher exposures of 250 and 750 mg/kg/day, but even at 60 mg/kg/day, one foetus exhibited a small lens malformation. Certain skeletal abnormalities were observed at 250 and 750 mg/kg/day, and incomplete or delayed ossification of certain bones (sternebrae and epiphyses) showed a dose-response relationship. Therefore, the study confirmed previous findings and indicated a lower NOAEL for the eye and skeletal malformations. Certain skeletal malformations (e.g. ribs) were observed at lower exposure levels but were not significant because, strangely, they were observed in the control group too.

All doses recorded higher incidences of 'retroesophageal right subclavian artery'[16] compared with both control and historical control data. Liver pathology of dams was observed at 60 mg/kg, and altered blood chemistry (high mean triglyceride and lower creatine levels) was observed from 30 mg/kg exposure onwards. The industry proposed a NOAEL of 60 mg/kg/day for maternal and developmental effects, a much lower threshold than the previous study.

A clear error of the study is that the TFA levels in the feed and water of the exposed animals were not taken into account. While TFA contamination was even detected in the control (up to 0.626 mg/L in the dams), this was not considered in the study conclusions, which could explain some of the skeletal malformations seen in this group. Moreover, the mechanistic part of the study did not provide insight into the way the eye malformations arise.

Another developmental rat study (OECD 414) was submitted by Bayer (see Annex nr. 3), but seems less relevant since it consists of a mixture of two PFAS substances (sulfonimide and potassium TFA). Based on exposure to this mixture (containing 14% KTFA), a NOAEL of 1000 mg/kg/day was claimed as no effects were observed.

Summary

The two developmental studies in rabbits provide clear and consistent evidence of serious foetal abnormalities, affecting both soft tissues (eyes) and skeletal structures. Industry proposes a NOAEL of 60 mg/kg/day, even though liver toxicity was already evident at that dose. At 30 mg/kg/day, altered blood chemistry was observed in the mother animals (dams), indicating systemic effects and suggesting that the NOAEL should be set even lower.

Crucially, similar toxic effects have also been documented in rats. Rather than acknowledging a cross-species pattern, industry dismisses the rat findings by claiming the results fall within "historical control data ranges." This deflection fails to address the consistency of effects across studies, reinforcing the conclusion that TFA's toxicity is not species-specific.

Moreover, a major methodological flaw casts doubt on the integrity of the studies' reporting: TFA levels in feed and water were never analysed during testing. This omission means that control animals may have been unknowingly exposed to TFA, potentially masking effects at low doses and suggesting that the true NOAEL could be lower than reported.

[16] In the 'normal' foetus three large arteries arise from the arch of the aorta: the brachiocephalic trunk (divided into the right common carotid artery and the right subclavian artery), the left common carotid artery, and the left subclavian artery. However, when the aberrant right subclavian artery variant is present, the brachiocephalic trunk is absent and four large arteries arise from the arch of the aorta: the right common carotid artery, the left common carotid artery, the left subclavian artery, and the final one with the most distal left sided origin, the right subclavian artery, also called the arteria lusoria. This vessel travels to the right forelimb, crossing the middle line of the body and usually passing behind the oesophagus.

Table 1: Summary of the industry development toxicity studies

Study	Species	Dose Levels (mg/kg/day)	Key findings	No-Observed-Adverse-Effect-Level (NOAEL)	Notes
1/ Prenatal Development Toxicity Study, Bayer, 2010 (OECD 414)	Rat	0 (control), 37.5, 75, 150	Eye malformations (one at 37.5; multiple at 150 mg/kg/day) Skeletal abnormalities (37.5; 150 mg/kg/day) Statistically significant ↑ in liver (+9.6%) and kidney (+5.6%) weights (150 mg/kg/day)	75 mg/kg/day for maternal toxicity150 mg/kg/day for embryo-foetal toxicity	Organ weight increases not considered adverse. foetal abnormalities considered within historical control data.
2/ Prenatal Development Toxicity Study, Bayer, 2021 (OECD 414)	Rabbit	0 (control), 180, 375, 750	Major eye malformations (folded retina, absent aqueous/vitreous humour) at 375 & 750; one at 180 mg/kg/day Skeletal abnormalities (375, 750 mg/kg/day) Heart/spine/kidney abnormalities (375, 750 mg/kg/day) ↓foetal/pup body weight (275, 650 mg/kg/day) ↑ liver weight & hypertrophy across all exposures	Bayer suggested that eye malformations at 180 mg/kg/day were within HCD range but did not propose a NOAEL based on the study .	Study report states NOAEL cannot be established.
3/ Prenatal 'mechanistic' Development Toxicity Study, TFA Task Force, 2024 (OECD 414)	Rabbit	0 (control), 30, 60, 250, 750	Major eye malformations (folded retina, absent aqueous/vitreous humour) at 250 & 750; small lens malformation at 60 mg/kg/day– Certain skeletal abnormalities (250, 750). Incomplete/delayed ossification (sternebrae, epiphyses) with dose response Rib malformations seen at low doses (also in controls)-Liver pathology (60, 250, 750 mg/kg/day) Retroesophageal subclavian artery anomalies (all doses) Blood chemistry alteration (from 30 mg/kg/day)	Proposed NOAEL of 60 mg/kg/day by the TFA task force despite abnormal effects even at 30/60 mg/kg/day: blood chemistry, liver histopathology, and one foetal eye issue	TFA detected in the control group (up to 0.626 mg/L in the dams). Feed/water contamination not considered. Study did not clarify the mechanism underlying the observed eye malformations.

2. Extended One-Generation Reproductive Toxicity Study

An **extended one-generation reproductive toxicity study** in rats (OECD 443) was also submitted by Bayer and Solvay in 2021 (see Annex 4), at the request of ECHA. The study focused on fertility and the endocrine system with doses of approximately 10, 50 and 250 mg/kg bw/day[17]. No adverse effects were reported by the industry, and the progression of the spermatogenic cycle was reported to be normal. Overall, according to the study report, *"reproductive performance showed no adverse effects of treatment"*[18].

However, by examining the study results closely, we find the following observed adverse effects that were not reported in the study conclusions:

- **Hormone disruption:** Thyroid hormone thyroxine (T4) dropped significantly in offspring at 50 (medium) and 250 mg/kg (high) in males, and at 250 mg/kg in females on 22 days of age (PND 22); T4 dropped in males at all exposures in 13 weeks of age Thyroid-Stimulating Hormone (TSH) increased in females at all doses on PND22, and in males at medium and high exposure.
- **Reproductive organs:** absolute weights of the testis were significantly reduced at high dose, as well as absolute epididymal weight in offspring. In parental males, a decrease in the weight of the epididymis and total sperm was observed in the high exposure group, along with a decrease in normal sperm and an increase in abnormal sperm (sperm parameters in parental animals were not examined in other exposure groups).
- **Organ weight:** Liver body weight ratio increased in both offspring males and females at high exposure (from 250 mg/kg); relative thyroid weight ratio decreased in males throughout all exposures and females at high exposure.
- **Sperm quality in offspring:** Lower testis weight and sperm counts were observed at the high dose of 250 mg/L (medium and low doses not tested); sperm motility was altered (Average Path Velocity decreased at all exposures 10, 50 and 250; Curvilinear Velocity decreased at 50 and 250 mg/kg); and abnormal sperm indicators increased at high dose (head abnormal & head flat).
- Changes in blood biochemistry were observed in parental animals (lower plasma glucose & non-esterified fatty acids; triglycerides, bilirubin, sodium, potassium and calcium alterations), as well as in offspring (alanine amino-transferase, bilirubin, glucose, cholesterol, triglycerides, sodium and calcium in males, potassium in females). Some of these effects were observed at all exposure levels.
- Some effects on the immune system were reported, where a decreasing trend was observed in the cells/spleen immunophenotyping parameters in offspring of both male and female across all treated groups
- Three out of 20 female foetuses developed eye abnormalities at high doses (rosettes/folds, retina), while foetuses at low and medium doses were not examined. Such abnormalities were not observed in high-dose males. Overall, although samples of eye tissue were collected across all groups, they were not analysed.

[17] The exact exposures were 0, 120, 600, 3000 ppm premating (9.71, 49.2 and 248 mg/kg/day for males and 10.26, 53.9 and 265 mg/kg/day for females) and 0, 60, 300, 1500 ppm during lactation (9.85, 47.5 and 233 mg/kg/day) and for offspring (male offspring: 0, 9.37, 47.3, 242 mg/kg bw/d; female offspring: 0, 9.83, 49.4, 248 mg/kg bw/day).

[18] 2021, Bayer-Solvay, page 16.

- Visceral abnormalities (including eyes) were not systematically examined; skeletal abnormalities were not examined at all.
- A specific cohort on immunotoxicity was not included.

Abnormalities in soft tissue and skeletal development were not evaluated. Based on the findings, the industry proposed a NOAEL of approximately 250 mg/kg/day.

Summary

Solvay-Bayer's 2021 extended one-generation study in rats claimed no adverse reproductive effects from TFA exposure. However, a closer look reveals overlooked signs of harm in offspring, including reduced thyroid hormone levels, weight of reproductive organs (testis and epididymis), liver changes, blood biochemistry alterations, and sperm abnormalities, even at lower exposures. Overall, eye malformations were not evaluated in parental animals or offspring at different stages. Some limited data are given for female and male offspring of 13 weeks at high exposure, where eye malformations were reported in females. These different effects were also identified by the dossier submitter (Germany competent Authority) in the course of hazard assessment by the Risk Assessment Committee at ECHA, and very recently by EFSA. It is important to note that skeletal malformations were not evaluated, and therefore, a comparison with the adverse effects observed in these tissues in the rabbit developmental toxicity study is not possible. Despite these gaps and findings, the industry proposed an unrealistically high NOAEL of 250 mg/kg/day.

Table 2: Summary of the Extended One-Generation Study

Study	Species	Dose Levels (mg/kg/day)	Key adverse findings	No-Observed-Adverse-Effect-Level (NOAEL)	Notes
Extended One-Generation Reproductive Toxicity Study, Bayer, 2021 (OECD 443)	Rat	approx. 0, 10, 50, 250	<u>Offspring effects</u>: Hormones: ↓ Thyroxine (T4) in males (50 & 250) and females (250) on Day 22; ↓T4 males (10, 50 & 250) at 13 weeks; ↑TSH females (10, 50, 250) Day 22; Organs: ↓Thyroid weight males (10, 50, 250) females (250); ↓ testis & epididymis (250); ↑ liver/body weight ratio male, female (250). Blood chemistry: ↑ alanine amino-transferase (250) & bilirubin (50, 250), ↓ glucose (10, 50, 250), ↓ cholesterol males (50, 250) & triglycerides males & non-esteridies fatty acids (10,50,250), ↑ Na^+ & ↓ Ca^{2+} in males, ↑ K^+ in females at all doses Offspring eye findings: 3/20 female foetuses at 250 showed retina malformations (rosettes/folds); no findings in males; lower doses not examined. Sperm quality in offspring: ↓ testis weight & sperm counts at 250; ↓ sperm motility (APV at 10/50/250; CLV at 50/250); ↑ abnormal sperm (head abnormal/head flat) at 250. Immunotoxicity: ↓ number of cells/spleen for different lymphocytes across all doses, males & females. <u>Parental effects</u>: ↓ weight of epididymis & total sperm (250), ↓ normal sperm & ↑ abnormal sperm (250). Blood chemistry: ↓ glucose (m,f, all exposures) & non-esterified fatty acids; altered Na^+, K^+, Ca^{2+}. Visceral & skeletal abnormalities not systematically examined.	3000 ppm (≈250 mg/kg/day)	Study focused on fertility & sperm endpoints; eye and skeletal developmental malformations largely unexamined.

3. Additional mechanistic studies

In 2024, alongside the new developmental toxicity study in rabbits, the TFA Task Force submitted a series of studies designed to further clarify the mode of action of TFA. These studies include:

- ***In vitro* CYP26 Binding Study:** An *in-vitro* study assessed TFA's potential to bind to human CYP26 (all-trans retinoic acid monoxygenase) in bactostomes (bacterial membranes). CYP26 is an enzyme which helps control levels of retinoic acid (a form of vitamin A and crucial for eye development). The hypothesis was that eye problems (folded retinas) in rabbits occurred via alterations in the CYP26 pathway, although this has not been proven. The result was negative, i.e. TFA does not act via human CYP26, and potentially neither via rabbit CYP26, due to the gene similarity. (see Annex, nr.5)

- ***In vitro* MCT1 Binding Study:** Another *in-vitro* study study examined TFA binding to the human protein Monocarboxylate transporter 1 (MCT1). MCT2 transports monocarboxylases through membranes, including lactate, a key molecule of metabolism acting as an energy source and cell regulator. Binding and transport were demonstrated at relatively high concentrations, which are much higher than levels humans would normally be exposed to (EC50 of 12 mM). (see Annex, nr.6)

- ***In vivo* Toxicokinetics and Clinical Observations in Rabbits:** This study looked at how TFA behaves in the body of rabbits and monitored certain health indicators, like lactate and vitamin A levels. It showed high internal plasma concentrations of TFA in non-pregnant female rabbits that were in the range or higher than the EC50 of the MCT1 assay. The TFA task force concluded that 750 mg/kg/day could be used for an *in vivo* embryo-foetal study. (see Annex, nr.7)

- **Toxicokinetic Mode-of-Action Study with sodium trifluroacetate:** A toxicokinetic-study in rabbits aimed at elucidating the mode of action for previously observed eye findings was reported. The toxicokinetic evaluation showed that sodium trifluoroacetate was metabolised to TFA, resulting in very high TFA internal exposure levels, with individual and mean Cmax levels (maximum blood concentration) of up to 20 mM and 16.5 mM, respectively. Both pregnant rabbits and foetuses had high plasma TFA levels. In pregnant rabbits, accumulation was observed at the 10 and 45 mg/kg/day dose levels after multiple repeated doses, but did not continue to rise at 180 and 750 mg/kg bw/d. Interestingly, though, TFA concentrations in the eyes of foetuses were higher than for the dams (parental) and showed a dose-response increase. Lactate, on the other hand, showed no fluctuation with exposure. Overall, TFA concentration in the plasma (blood) of rabbits was higher than in the eyes. (see Annex, nr.8)

- **Read-across from TFA:** A NOAEL is derived of about 210 mg/kg/day. (see Annex, nr.10)

Industry's failed attempt to disprove developmental toxicity evidence

Along with the additional studies, the Industry Task Force provided a paper entitled *"Position Paper to explain the Mode of action for observed foetal eye findings in rabbits"*, presenting a hypothesis as to why the observed eye malformations are rabbit-specific. However, this argument is speculative and not supported by the evidence.

In vitro studies indicate that TFA binds to the lactate receptor and activates it, mimicking the effects of lactate. In its position paper, industry proposes that TFA competes with the lactate transport, causing alterations in pH homeostasis and energy supply in the ciliary body epithelium of the eye, which is responsible for producing aqueous humour and maintaining the eye's internal environment. This disruption may impair ion transport, reducing aqueous and vitreous humour secretion into the eye's chamber and contributing to eye malformations in rabbit foetuses.

The industry TFA Task Force suggests an internal TFA threshold level, that when it is reached TFA binds to MCT transporters 1 in ocular/eye tissues: 12mM. Therefore at that concentration TFA competes with natural lactate and blocks its action. To prove this, they provided an *in vitro* study (Annex Study 6) showing that TFA binds the human MCT1, similarly to lactate, however without proving any competition. The industry consortium also provided a toxicokinetic study in rabbits, measuring the levels of TFA in the blood of the rabbits, which actually shows that the levels of TFA in the blood of animals are increasing, but do not reach this threshold, apart from those in the high exposure group (750 mg/kg).

Therefore, in the extensive analysis by the Task Force, their hypothesis is not supported by the evidence. Significant eye malformations in rabbits' offspring were observed in two different studies at concentrations of 180, 250, 375 and 750 mg/kg bw/day. The internal exposure to TFA in plasma exceeded the trigger threshold mentioned above of EC50 for MCT of 12mM only in dams from the highest exposure group (750 mg/kg). In all the other exposures where rabbits developed eye malformations, the internal TFA was below this hypothetical threshold. Therefore, even the evidence from the industry's own studies does not support this hypothesis and should be dismissed.

Moreover, the Task Force misleadingly mentions that *"comparable findings were not observed in a developmental toxicity study in rats"* and refers to the two industry studies provided, where either eye samples were taken but not fully reported (EOGRTS, 2021) or eye malformations were not measured at all (2010). In fact we now know that there was a rat study (90-d repeated dose toxicity study, OECD TG 408) by Bayer itself from 2007, where eye malformations were also observed in males from high dose exposure (876 mg/kg). Another study from 2019 (1-year repeated dose toxicity study, OECD TG 452), also reports eye malformations in exposed animals at much lower doses *"Cornea opacity, cataract, ocular discharge, periorbital swelling, iris synechia, retinal vessel tortuosity, fundus hyperreflectivity and incomplete pupil dilation post-mydriatic were observed in some animals during the dosing and recovery phase exams."* Unfortunately, since the study is unpublished, the specific details are unknown to us. The Task Force failed to mention any of these studies that clearly indicate that the eye is a target organ in adult rats after chronic oral exposure and therefore prove that TFA may cause eye malformations in rats too.

Our analysis exposes that the Task Force companies have provided scientifically unfounded arguments attempting to mislead the TFA toxicity assessment that the eye malformations caused by TFA exposure in foetuses are rabbit-specific and undermine the developmental toxicity potential of TFA.

In addition, they develop no arguments about the observed skeletal malformations, which were also observed, but to a lesser extent in rats. Finally, the changes in thyroid parameters, reproductive organs and sperm quality observed in rats are not discussed at all.

Summary

Industry attempted to downplay TFA's eye toxicity by proposing a rabbit-specific mechanism tied to lactate transport disruption. However, malformations occurred at TFA levels below the proposed hypothetical threshold they calculated to trigger such effects, undermining their own argument. Moreover, evidence from multiple rat studies, including data from Bayer, shows eye malformations in adult rats as well. This suggests the eye is a general target organ, not limited to rabbits. Industry also failed to address skeletal malformations seen across species, reinforcing concerns about broader developmental toxicity.

Independent studies

Independent studies on the toxicity of TFA are limited. One notable 1984-study[19] on pregnant mice shows that inhalation of halothane, an anaesthetic, with TFA as a metabolite, resulted in high activity of the metabolites in the neuroepithelium of the embryo in early gestation and high concentrations of radioactivity in amniotic fluid (and the ocular fluids of adults) still prevailing at 24 hours after inhalation. Industry must have been aware of this study and studied these effects.

A similar 1996-study in rats[20] demonstrated changes in liver biochemistry of newborns at 75 mg/kg/day, while TFA caused a functional deficit of the proximal tubule in newborns. The authors show that the effects are transient.

"Safe" exposure levels and reference values

In this section, we examine how so-called "safe" exposure levels for TFA are defined. From Acceptable Daily Intakes (ADIs) to drinking water limits, we present the toxicological reference values adopted so far by different authorities and the scientific rationale behind them. For a detailed analysis of the underlying methodologies and of whether these values truly safeguard the most vulnerable populations, we refer readers to the report *TFA: Générations Futures alerte sur la gestion des pouvoirs publics*.

We also shed light on **how industry actors are actively lobbying for the adoption of comparatively high drinking water limits.**

EFSA

In its 2014 opinion on MRL-setting for saflufenacil, EFSA established an Acceptable Daily Intake (ADI) for TFA of 0.05 mg/kg bw/day, based on a 90-day rat study (Bayer, 2007). The critical effect was increased liver weight, together with hepatocellular hypertrophy linked to peroxisome proliferation (also necrosis and increased alanine aminotransferase), applying an uncertainty factor of 200 for extrapolation to chronic exposure. The NOAEL (target organ: liver), as proposed by Bayer in its position paper, was 10 mg/kg bw/day for males and 12 mg/kg bw/day for females (increased liver weight due to hepatocellular hypertrophy driven by peroxisome proliferation).

EFSA's ADI was established solely for the purpose of pesticide risk assessment. However, no EU-wide drinking water limit for TFA has been set by European regulators. In the absence of such a standard, several Member States have introduced their own national guidance values for drinking water, building on different methodologies.

[19] B R Danielsson, et al, Accumulation in murine amniotic fluid of halothane and its metabolites, Acta Pharmacol Toxicol (Copenh) 1984 Nov;55(5):410-7

[20] A. M. Saillenfait, et al, Postnatal Hepatic and Renal Consequences of in Utero Exposure to Halothane or its Oxidative Metabolite Trifluoroacetic Acid in the Rat, JOURNAL OF APPLIED TOXICOLOGY, VOL. 17(1), 1–8 (1996).

In July 2024, the European Commission requested EFSA to issue an EFSA statement and review the recommended toxicological reference values, i.e. acceptable daily intake (ADI) and acute reference dose (ARfD), for TFA.

In line with its mandate, EFSA is currently proposing an ADI at 0.03 mg/kg bw per day based on a NOAEL of 8.65 mg/kg bw per day from the EOGRTS together with an uncertainty factor (UF) of 300.

Germany (2020)

Germany derived a reference value from a 52-week drinking-water study in rats (Solvay, 2019). Based on observed liver damage and elevated alanine aminotransferase levels, the German Environment Agency (UBA) established an ADI of 0.018 mg/kg bw/day. This was based on a NOAEL of 1.8 mg/kg bw/day, applying a standard safety factor of 100 to account for differences within and between species. However, no additional safety factor was included to reflect the existing uncertainties around TFA's full toxicological profile.

Based on the ADI, and assuming that a consumer weighs on average 70 kg, drinks 2 litres of water per day, and considering that water accounts for 10% of overall exposure to TFA, UBA calculated a drinking water value of 63 µg/L, rounded down to **60 µg/L**. This approach is not protective, particularly with regard to women and children, who are effectively excluded from the assessment by using an average body weight of 70 kg.

Luxembourg (2024)

Luxembourg's Health Directorate (DISA) followed a similar approach to that of UBA, adopting the same ADI of 0.018 mg/kg bw/day for TFA. However, like UBA, they did not apply any additional safety factors to account for the significant uncertainties due to limited toxicity data.

Where Luxembourg differed was in how it calculated the drinking water guideline. Instead of assuming an average adult weighing 70 kg, DISA based its calculation on a more vulnerable population: a 5 kg infant consuming 0.75 litres of water per day. As a result, the derived drinking water value is **12 µg/L**, which is five times lower (and more protective) than the 60 µg/L set by Germany's UBA.

The Netherlands (2023)

The Dutch competent authority (RIVM) has been critical of the UBA calculation, arguing that a clear dose–response relationship is required. According to RIVM, such a relationship is absent for ALT but present for bilirubin and liver weight. For drinking water, the RIVM applies the EFSA-derived Tolerable Weekly Intake (TWI) of 4.4 ng/kg bw/week (immune effects in breast-fed children) for four PFAS with evidence of immune effects. From this TWI, they derived a drinking-water standard of 4.4 ng/L for PFOA. Given similar effects (bilirubin, liver weight), TFA is included in this group, assuming comparable immune effects. Because TFA shows lower potency (based on liver weight; Bayer, 2007), this results in a drinking-water standard of **2.2 µg/L**.

In the Netherlands, the total PFAS burden (in PFOA equivalents) is calculated to be, on average, above the EFSA TWI standard (low-bound scenario), with the 95th percentile reaching three times the TWI (also LB). Only the Netherlands has considered children's exposure in their calculations.

Flanders (Belgium) (2024)

Flanders' research institute VITO based its safety assessment on two studies, combining data from Bayer's 2007 90-day study and Solvay's 2019 one-year study, instead of relying on a single study. Through statistical analysis of the combined data, VITO found a significant drop in bilirubin levels in rats exposed to TFA versus control animals. This decrease in bilirubin was identified as the key effect used to establish safe exposure limits. The dose causing at least a 30% decrease in bilirubin (known as the BMDL30) was estimated at 5.2 mg/kg body weight per day.
What sets VITO apart is its choice of an additional safety factor of 10, applied in addition to the standard uncertainty factors. This extra precaution was added to address gaps in current data, particularly regarding immunotoxicity, reproductive effects, and developmental toxicity of TFA.

Finally, when translating the ADI into a drinking water guideline, VITO assumed a standard adult weight of 60 kg. Assuming a daily water consumption of 2 litres and considering that water accounts for 20% of total TFA exposure, this results in a drinking water guideline value of **15.6 µg/L**. Unlike Luxembourg, this calculation did not specifically account for children or more vulnerable groups, which means the resulting water quality value may be less protective for these populations.

Table 3: Guidance health-based value derived for TFA in drinking water

Germany (2020)	60 µg/L
The Netherlands (2023)	2200 ng/L (2.2 µg/L)
Luxembourg (2024)	12 µg/L
Flanders (2024)	15.6 µg/L

Industry's proposal

The industry Task Force welcomes EFSA's approach from 2014, with a NOAEL of 10 mg/kg based on liver toxicity and a final ADI of 0.05 mg/kg, but they are critical of both Germany and the Netherlands. For Germany (UBA), they argue that the NOAEL based on changes in liver biochemistry in the 1-year rat study is "not appropriate", claiming such effects are not considered adverse. For the Netherlands, they criticise the inclusion of TFA as a PFAS, describing the approach as of "low value" and unjustified, and assert that TFA already has a sufficient dataset. Nevertheless, key long-term studies, such as carcinogenicity and certain genotoxicity studies, are still lacking.

Remarkably, the Task Force presents its own calculations for a health-based drinking-water limit. Based on the EFSA ADI, they propose a health-based value of **294 µg/L**. This calculation uses only adult exposure data, which leads to a much higher figure, as children consume proportionally more water and food relative to their body weight. Their proposed value is **133 times higher** than the drinking-water safety value endorsed by the Netherlands. The companies also overlook the fact that TFA is detected in drinking water across Europe, meaning that young children and other vulnerable groups are likewise exposed.

Summary

Across Europe, derived values for TFA in drinking water span more than an order of magnitude: from 2.2 µg/L in the Netherlands, to 12 µg/L in Luxembourg, 15.6 µg/L in Flanders, and 60 µg/L in Germany. By contrast, the industry Task Force has proposed a value of 294 µg/L, more than 100 times higher than the most protective national standards.

Conclusion

TFA stands at the heart of Europe's PFAS problem. As both a direct pollutant and the breakdown product of PFAS pesticides and other chemicals, it has become the most widespread PFAS in the environment. TFA contaminates groundwater, drinking water, and the food chain, creating a long-term burden for European societies.

The evidence assembled in this report demonstrates that:

- **TFA is harmful.** Different studies consistently show adverse effects, including developmental toxicity, thyroid disruption, liver damage, effects on immune system and impacts on sperm parameters. Many of these findings are observed in more than one study or species and must not be considered isolated or incidental.

- **The industry has delayed and distorted the assessment procedure.** For decades, companies selectively framed data, avoided conducting critical long-term studies until legally compelled, and advanced speculative arguments to downplay adverse effects. Their proposed drinking water value of 294 µg/L, based only on adult exposure and significant toxicological data gaps, is more than 100 times higher than the most protective national standards and disregards the protection of vulnerable groups, including babies and children.

- **Regulators have failed to act in time.** Early warnings date back to 1998, yet approvals of PFAS pesticides continued, leading to predictable contamination of groundwater and food.

As the EU reassesses TFA's hazard classification and health-based reference values, several conclusions follow:

- **The precautionary principle must apply.** The persistence, mobility, and toxicity of TFA mean that waiting for further proof will only deepen irreversible contamination. Regulatory decisions should reflect worst-case exposure scenarios and explicitly protect vulnerable populations.

- **Independence of assessments must be guaranteed.** Strong rules are needed to ensure that risk assessments are independent from industry influence, both at the EU and Member State level. Transparency, rigorous standards, and independent scientific expertise are essential to avoid regulatory capture, biased assessments, and the prioritisation of corporate interests over public health.

- **Health-based values must be protective.** National assessments show that protective drinking water values are in the low µg/L range (2.2–15.6 µg/L). Any higher limits, such as those promoted by industry, would normalise widespread exposure and fail to safeguard children and sensitive groups.

- **PFAS pesticides must be phased out.** Continued approval of active substances that degrade into TFA is incompatible with the legal requirement to prevent groundwater contamination and protect human health.

Twenty-five years after the first warnings, Europe has the opportunity, and the obligation, to prevent further TFA contamination, uphold the law, and secure safe water for present and future generations.

Annexes - notes on the studies

1. Bayer 2021 study embryo-foetal development in rabbit, New Zealand White strain because of available HCD - study Number 8437242 - (OECD 414), Renault R., Covance Laboratories, (2021). Study plan JUN 2020, draft report AUG 2021.
Four times 24 rabbits, control and three doses, 180, 375 and 750 mg/kg/day (four groups).
Females treated from day 6 – 28 after mating. Kill day 29.
(note: there was no analysis of TFA content in feed & water of controls).

Main findings: Litter, 375/750 mg/kg/day: major eye abnormalities, multiple folded retina (disparity in growth of the inner and outer retinal layers) and absent aqueous vitreous humour (bodily fluids located in the eyes). At 180 mg/kg also one foetus with microphthalmia (one or two small eyes), multiple folded retina and absent aqueous vitreous humour.
180 mg/kg/day-finding: within HCD but treatment-relation cannot be ruled out.

In addition at 375/750 mg/kg/day, there were low incidences of major/minor abnormalities affecting heart and major blood vessels: transposition of ascending aorta/pulmonary trunk (embryological discordance between the aorta and pulmonary trunk); narrow/dilated ascending aorta and pulmonary trunk (significant aortic valvular insufficiency); double outlet ventricle(s) and ventricular septal defects (a hole between the lower heart chambers); multiple cervical/thoracic/lumbar/caudal vertebral and rib abnormalities (misshaped spine and ribs); with associated minor abnormalities affecting the vertebrae, ribs, sternum and costal cartilages); kidneys: absent/displaced/fused.
Bayer comments that the relation of these effects with treatment is unlikely and within HCD.

Also, there was an increased incidence of the minor findings 20 thoracolumbar vertebrae, unossified 5th sternebrae and epiphyses, incompletely ossified cervical/thoracic vertebrae and metacarpals and forepaw flexure compared to concurrent control.
Bayer: outside or within HCD, not adverse but may-be treatment related.

Foetal eyes: examination revealed slight to marked retinal folds in the majority (5 of 8) of foetuses whose dams were administered 375 mg/kg/day and in all foetuses (11 of 11) whose dams were administered 750 mg/kg/day and demonstrated a relationship to dose in terms of relative group incidences. Marked retinal folds were seen in the one foetus whose dam was administered 180 mg/kg/day. Lens degeneration was seen in several foetuses (6 of 20), and haemorrhage in the vitreous chamber was seen in a few foetuses (2 of 20), that had retinal folds. The majority of the foetuses that had retinal folds also had absence of aqueous and/or vitreous humour (13 of 20, generally bilateral) reported at macroscopic examination.

2. Developmental Toxicity Study in Rats with TFA, 2010 in US, report nr. 09-4352 (provided by Bayer). OECD 414. Rat Strain: Sprague-Dawley, Huntingdon Life Sciences.
The study was designed to assess any maternal and/or embryofoetal toxicity when TFA was administered daily by gavage to rats from implantation to the day prior to expected parturition. Groups of 22 timed-mated Sprague-Dawley rats were administered TFA in distilled water once daily by gavage at dosages of 0 (control), 37.5, 75 or 150 mg/kg bw/day at a dose volume of 5 mL/kg bw, on gestation days (GD) 6 to 19 inclusive.

At termination on GD 20: Gross necropsy, liver and kidney weights, gravid uterus weight, counts of corpora lutea and implantations (live, early or late deaths), foetal weights, placental weights, then subsequent examination for foetal abnormalities and variants (soft tissue and skeletal).

Outcome: There were no treatment related effects on the pregnancies in terms of post-implantation loss, foetal or placental weight, or the incidence of foetal abnormalities and variants, including skeletal ossification status (NOAEL 150 mg/kg).

Data on foetuses and litter:

		Fetuses				Litters			
	Group	1	2	3	4	1	2	3	4
Observation	**External Examinations**	122	141	145	125	21	22	22	21
Testis, displaced, cranial			1	1			1	1	
Testis, displaced, medial		2	11	7	2	2	9	5	2
Testis, small					1				1
Umbilical artery, left		1	2	2	3	1	2	2	3
Brain									
3rd ventricle, hemorrhage(s)			1				1		
4th ventricle, dilated			1				1		
Cerebellum, subdural, hemorrhage(s)		2	8	5	3	2	5	4	3
Cerebral hemisphere, mid brain, hemorrhage between		1		2	1	1		2	1
Cerebral hemisphere, hemorrhage(s)		1		1		1		1	
Cerebral hemisphere, subdural, hemorrhage(s)		3			3	2			3
Medulla oblongata, subdural, hemorrhage(s)				2	1			1	1
Cervical									
Thymus gland, partially undescended		2	3	3	5	2	3	2	4
Eye									
Lens, oval			1		2		1		1
Orbital sinus, dilated				1				1	
Orbital sinus, hemorrhage(s)		1	4	1	1	1	3	1	1
Retina, folded					1				1
Head									
Cranial region, subcutaneous, hemorrhage(s)			2				1		
Head (general), hemorrhage(s), subcutaneous		1	6	1		1	3	1	
Jaw, lower, subcutaneous, hemorrhage(s)			4		1		3		1
Vibrissae region, subcutaneous, hemorrhage(s)			1		1		1		1
Posterior, subcutaneous, hemorrhage(s)		1	1		2	1	1		2

2. Developmental Toxicity Study in Rats with TFA, 2010 in US, report nr. 09-4352 (provided by Bayer). OECD 414. Rat Strain: Sprague-Dawley, Huntingdon Life Sciences.
The study was designed to assess any maternal and/or embryofoetal toxicity when TFA was administered daily by gavage to rats from implantation to the day prior to expected parturition. Groups of 22 timed-mated Sprague-Dawley rats were administered TFA in distilled water once daily by gavage at dosages of 0 (control), 37.5, 75 or 150 mg/kg bw/day at a dose volume of 5 mL/kg bw, on gestation days (GD) 6 to 19 inclusive.

At termination on GD 20: Gross necropsy, liver and kidney weights, gravid uterus weight, counts of corpora lutea and implantations (live, early or late deaths), foetal weights, placental weights, then subsequent examination for foetal abnormalities and variants (soft tissue and skeletal).

Outcome: There were no treatment related effects on the pregnancies in terms of post-implantation loss, foetal or placental weight, or the incidence of foetal abnormalities and variants, including skeletal ossification status (NOAEL 150 mg/kg).

Abnormalities of soft tissue and skeletal were observed but claimed to be within HCD; one case on “eye- retina, bilateral, folded” and in highest dose clear skeletal problems.

Data on foetuses and litter:

		Fetuses				Litters			
	Group	1	2	3	4	1	2	3	4
Observation	**External Examinations**	122	141	145	125	21	22	22	21
Testis, displaced, cranial			1	1			1	1	
Testis, displaced, medial		2	11	7	2	2	9	5	2
Testis, small					1				1
Umbilical artery, left		1	2	2	3	1	2	2	3
Brain									
3rd ventricle, hemorrhage(s)			1				1		
4th ventricle, dilated			1				1		
Cerebellum, subdural, hemorrhage(s)		2	8	5	3	2	5	4	3
Cerebral hemisphere, mid brain, hemorrhage between		1		2	1	1		2	1
Cerebral hemisphere, hemorrhage(s)		1		1		1		1	
Cerebral hemisphere, subdural, hemorrhage(s)		3			3	2			3
Medulla oblongata, subdural, hemorrhage(s)				2	1			1	1
Cervical									
Thymus gland, partially undescended		2	3	3	5	2	3	2	4
Eye									
Lens, oval			1		2		1		1
Orbital sinus, dilated				1				1	
Orbital sinus, hemorrhage(s)		1	4	1	1	1	3	1	1
Retina, folded					1				1
Head									
Cranial region, subcutaneous, hemorrhage(s)			2				1		
Head (general), hemorrhage(s), subcutaneous		1	6	1		1	3	1	
Jaw, lower, subcutaneous, hemorrhage(s)			4		1		3		1
Vibrissae region, subcutaneous, hemorrhage(s)			1		1		1		1
Posterior, subcutaneous, hemorrhage(s)		1	1		2	1	1		2

Slight, test article-related, statistically-significant increases in liver and kidney weights were observed at 150 mg/kg/day (+9.6% and 5.6%, respectively). These increased liver and kidney weights were not considered adverse. Industry: due to the non-adverse, test article-related organ weight increases, the maternal and embryo-foetal no-observed-effect-levels (NOEL) were established at 75 mg/kg/day (maternal).

3. Developmental Toxicity Study in Rats with Reaction Mass, 2020, report nr. G1963B0020 – (provided by Bayer)
OECD 414, Solvay, China. Rat Strain: Sprague-Dawley.
Mated females (27/group) were treated once daily with the Reaction mass of TFSK/TFAK (Reaction mass of potassium trifluoroacetate and potassium Trifluoromethanesulphinate) by oral gavage at dose levels of 100, 300, and 1000 mg active ingredient/kg bw/day during gestation (from Gestation Day 5 to 19). A control group of 27 females receiving the vehicle (water) was included in the study. Approximately half of the foetuses were examined for soft tissue abnormalities. The other half was examined for skeletal abnormalities and ossification state.

Outcome: No adverse effect attributable to treatment was observed across all groups with respect to external, soft-tissue and skeletal malformations or variations. Based on these results, the No-Observed-Adverse-Effect-Level (NOAEL) of the Reaction mass of TFSK/TFAK for maternal and embryo-foetal development toxicity in rats was considered to be 1000 mg/kg/day.

Questionable relevance since mixture of PFAS.

4. Extended 1-Generation Reproductive Toxicity Study in Rats with TFA – OCT 20 – May 21, Renault. R., 2021. Report nr. 8437241 (provided by Bayer).
OECD 443. Han Wistar Rat, Labcorp Laboratories.
In the F0 generation, three groups of 25 rats/sex received Sodium Trifluoracetate at dietary concentrations of 120, 600 or 3000 ppm, corresponding to nominal dose levels of ca. 10, 50 and 250 mg/kg bw/day, for 10 weeks premating, gestation and lactation. To compensate for the higher food intake during lactation, dietary concentrations were reduced in females to 60, 300 and 1500 ppm during lactation. The F1 animals consisted of 2 cohorts from weaning and were treated with the test item directly from weaning (Day 21 of age) up to 13 and 14 weeks, respectively. Day 21 to nominal Day 35 of age at dietary concentrations of 60, 300 or 1500 ppm and from Day 35 of age to their scheduled termination at dietary concentrations of 10, 600 or 3000 ppm.

Criteria for evaluation included viability (morbidity/mortality), clinical observations, body weight, food consumption, oestrus cycle, cohabitation duration, sperm analysis, fertility data, gestation duration, parturition observation, litter evaluation, litter data, development landmarks, clinical pathology (haematology, serum chemistry, urinalyses), thyroid hormone analysis (TSH and T4), spleen lymphocyte subpopulations, gross (necropsy) evaluation, organ weight, and histopathological evaluation (F1 Cohort 1B animals were not examined microscopically).

Outcome: Histopathological examination of the Cohort 1A showed that the majority exhibited normal progression of the spermatogenic cycle, and the expected cell associations and proportions in the various stages of spermatogenesis were present. Tubular degeneration and atrophy occurred at a low incidence in both treated and control males but was considered unrelated to treatment as the incidence was similar across the groups (NOAEL 3000/1500 ppm (approximating 242 -265 mg/kg/day)). Soft tissue and skeleton abnormalities not part of study.

5. In vitro study on CYP26 binding (TFA taskforce, BASF BAYER CHEMOURS CORTEVA HONEYWELL RHODIA SYNGENTA), submitted FEB 24, conducted in 2022/2023/2024 (in vivo).
Model: Recombinant human CYP26B1LR Bactosomes
Study on endocrine activity on CYP26 (all-trans retinoic acid monoxygenase), an enzyme in the retinoic acid pathway. Outcome negative.
The recovery of TFA was far lower than control, around 50% vs 75%.

6. In vitro MCT1 binding (TFA taskforce)
TFA though was a substrate of Monocarboxylate transporter 1 (MCT1), competing for normal substrate lactate. Model: DLD-1 human colorectal adenocarcinoma cells. A concentration range of 1 to 150 mM TFA was used.
TFA was shown to be transported with an EC50 of 12 mM by the human MCT1.

7. Proof of the concept study in the rabbit by Oral Gavage administration. **In vivo toxicokinetic and clinical observations (lactate, vit.A) (TFA taskforce, 2024) –**
Six rabbits exposed to 750 mg/kg bw/day and 6 controls.

No test of feed and water on TFA content in control.

Blood chemistry, high and low values of constituents.
At 750 mg/kg bw/d, very high plasma concentrations of trifluoroacetic acid (TFAA)) were measured, with individual Cmax values of 16.1 and 18.3 mM on day 1 and 13, respectively. Mean cmax values were 14.7 and 16.6 mM on day 1 and 13, respectively. Though no accumulation.
In vivo study showed very high internal exposure concentrations measured as trifluoroacetic acid (TFAA) in maternal and foetuses that were in the range or higher than the EC50 of the MCT1 assay. High TFAA levels were also determined in aqueous/vitreous humour samples of pregnant rabbits and foetuses.
TFA taskforce conclusion: 750 mg/kg/day can be used for an in-vivo embryo-foetal study.

8. Toxicokinetic (TK)-study in pregnant NZW rabbits (TFA taskforce).
Objective: the best design of the mechanistic developmental study (including dose levels) to elucidate the mode of action for previously observed eye findings.
No guideline study.
Four groups of six pregnant female NZW rabbits received Sodium Trifluoroacetate at doses of 10, 45, 180 or 750 mg/kg/day by oral gavage administration, from Day 6 to 28 after mating. Plus control group, Kill day 29.
Blood chemistry examination showed treatment-related changes at 750 mg/kg bw/d with low mean bile acids (-60% of Controls, $p<0.05$), high mean plasma urea (+28% of Controls; $p<0.05$), high mean cholesterol (+55% of Controls, $p<0.05$) and high mean triglyceride concentration (+74% of Controls, $p<0.05$). 3DHY was increased at all sodium trifluoroacetate levels, up to +227% compared to controls. NEFA were decreased at 45 mg/kg bw/d and above. Toxicokinetic evaluation showed very high internal exposure levels, with individual and mean Cmax levels of up to 20 mM and 16.5 mM, respectively. Accumulation of sodium trifluoroacetate was observed at the 10 and 45 mg/kg/day dose levels after multiple doses in pregnant rabbits, but not at 180 and 750 mg/kg bw/d. The levels of TFA in foetal samples were higher compared to the concentrations in eyes of the respective dams. There was no treatment-related effect on reproductive and litter parameters.

Each foetus was only externally examined. Only Aqueous/vitreous humour samples were taken.

9. Mechanistic Developmental Toxicity study (TFA taskforce) - Study No 8485229 - OECD 414- necropsy August 2021 – August 2022, Labcorp Laboratories.
The purpose of this study was to assess the influence of Sodium Trifluoroacetate (TFA-Na) on embryo-foetal survival and development when administered during the organogenesis and foetal growth phases of pregnancy in the NZW Rabbit. Additional investigations were included with the aim to assess maternal effects and examine potential mechanisms for the foetal eye pathology reported in Study 8437242. Four groups of 30 time-mated females received TFA-Na at doses of 30, 60, 250, or 750 mg/kg/day by oral gavage administration, from gestation day (GD) 6 to 28. Additional 8 time-mated females per dose were assigned to satellite phase for evaluation of toxicokinetics, lactate profile, clinical chemistry and blood pH investigations. These animals were treated from GD 6 to 20. Kill day 29. All live main phase foetuses were examined macroscopically at necropsy and subsequently by detailed internal visceral examination of the head or skeletal examination. At least one of the two lower dose levels of 30 and 60 mg/kg bw/day is expected to cause no foetal eye findings and thus provide a NOAEL. The dose levels should also enable to establish a dose concordance for the mechanism of action behind the foetal eye findings. They pool the samples generally to an average.

Liver pathology, altered metabolism and increased kidney weight were amongst the changes in female rabbits. Dose-related incidences of minimal to moderate bile duct hyperplasia were observed at 60, 250 or 750 mg/kg/day.

Detailed foetal examination revealed an increased incidence of major foetal abnormalities at 250 or 750 mg/kg/day; these abnormalities mainly affected the eye and showed consistency and/or exceeded the historical control range. Based on the maternal liver pathology showing bile duct hyperplasia/fibrosis, occurring in conjunction with other liver lesions, and the major foetal abnormalities at 250 and 750 mg/kg/day, 60 mg/kg/day was considered to be the maternal and foetal no observed adverse effect level (NOAEL).
Dose levels of Control, 30, 60, 250, or 750 mg/kg/day (groups 1-5), recorded incidences of 'retroesophageal right subclavian artery', which were higher - compared with both concurrent control and the historical control data ranges in terms of foetal and litter incidences at 250 and 750 mg/kg/day. Industry tries to change these malformations into simple 'variations' (i.e. non-adverse) in its position paper[21].

Outcome. In control: On GD 6 individual TFAA concentrations in plasma were in the range of 0.232 to 0.527 mg/L and 0.275 to 0.626, at 2 and 24 hours after dosing, respectively. On GD19 individual TFAA concentrations ranged from 1.41 to 57 mg/L and 1.17 to 53.6 mg/L, at 2 and 24 hours, respectively. On GD 28, two hours post dose, individual TFAA concentrations in main animals ranged from 0.45 to 2.69 mg/L. Although trifluoroacetate is endogenously present, especially some higher control values cannot be explained by this. However, since the control levels are far below the concentrations measured in the treated groups, they have no impact on the conclusions of this study (conclusion Taskforce).

TFA in control up to 57 mg/L in plasma is very strange. For comparison, exposed dams have around 300 mg/L.

Blood chemistry dams: Females receiving 30 or 60 mg/kg/day also showed high mean triglyceride levels on GD19 when compared with Controls ($p<0.05$).
Histology dams: At termination on GD 29 treatment related findings were observed in the liver at 60 mg/kg bw/d and above.

Exposure 750/250 grossly similar foetal damage as the study before (Study 8437242). At 60mg/kg/day (group 3) there was also one foetus in one litter with an incidence of small lens. Even in the control (group 1) there were skeletal abnormalities (see below).

Thus, dams at 30 and 60 mg/kg bw/d (group 2 and 3) had abnormal blood chemistry, at 60 mg/kg (group 3) liver histology problems, and at 60 (group 3) a foetus with eye problems.

[21]Although classified as a malformation by the conducting laboratory there is evidence to suggest that retroesophageal right subclavian artery does not adversely affect health or survival and that this finding should be considered a variation, reducing considerably its toxicological significance.

Table 36: Summary of fetal findings and laboratory Historical Control Data (HCD)

Historical control information: Rabbit/New Zealand White [redacted]
Necropsy date range August 2021 – August 2022
Number of studies 12

	Fetuses					Litters					HCD Range#	
Group	1	2	3	4	5	1	2	3	4	5		
Number Examined	209	244	219	193	178	28	29	28	28	28	2128	270
Total Number Affected	4	1	3	11	25	4	1	3	9	14		
Head												
Visceral												
Multiple folded retina	0	0	0	2	12	0	0	0	2	9	0-0	0-0
Absent aqueous/vitreous humour	0	0	0	1	8	0	0	0	1	7	0-0	0-0
Haemorrhage(s) aqueous/vitreous humour	0	0	0	0	5	0	0	0	0	5	0-0	0-0
Misshapen lens	0	0	0	0	2	0	0	0	0	2	0-0	0-0
Small lens	0	0	1	0	0	0	0	1	0	0		
Folded retina	0	0	0	0	1	0	0	0	0	1	0-0	0-0
Cervical/Thoracic											0-0.7	0-4.4
Skeletal												
Sternebrae, fused/partially fused	1	0	3	3	3	1	0	2	3	3	0-3.1	0-18.2
Sternebrae, bipartite ossified	0	0	0	0	3	0	0	0	0	3	0-1.1	0-5.0
Sternebrae, misaligned ossification sites	0	0	0	0	2	0	0	0	0	2	0-1.7	0-10
Sternebrae, misaligned hemicentres	1	0	0	0	3	1	0	0	0	3	0-1.5	0-9.5
Fused/partially fused ribs	0	1	0	1	6	0	1	0	1	3	0-0.5	0-4.6
Visceral												
Retroesophageal right subclavian artery	1	0	1	5	13	1	0	1	4	8	0-2.0	0.8.7

Note: Individual fetuses/litters may occur in more than one category.
Historical control information:(mean group fetal percentage / Percentage of litters affected) (need to be updated when final report is available)

10. Read-across from Sodium trifluoroacetate (ECHA dossier).

Based on read-across from Sodium trifluoroacetate, the NOAEL of trifluoroacetic acid will also be 3000/1500 ppm (corresponding to approximating 203 - 222 mg TFA/kg/day) for both reproductive performance/offspring development and for general systemic toxicity.

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Pesticide Action Network (PAN) Europe is a science-based organisation bringing together more than 50 consumer, public health and environmental organisations, trade unions, women's groups and farmer associations from across Europe. We work to eliminate the dependency on pesticides and promote alternatives that work instead of fighting against nature.

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