eurofins

WHITE PAPER

Trifluoroacetic acid (TFA) and trifluoromethane sulphonic acid (TFMS) in juice and fruit/vegetable purees

Patrick van Hees[1,2], Patrik Karlsson[1], Ludmila Bucuricova[1], Helena Olsman[1], Leo Yeung[2]

[1]PFAS Competence Centre, Eurofins Food & Feed Testing Sweden AB, [2]Man-Technology-Environment (MTM) Research Centre, Örebro University, Sweden

Summary

Trifluoroacetic acid (TFA) and trifluoromethane sulphonic acid (TFMS) were determined in juices and drinks (n=37) together with fruit/vegetable purees (n=9) for small children. The results showed elevated levels of TFA, especially, in orange juice and in some hand-squeezed oranges. Considering samples above the reporting limit, an average TFA level of 34 000 ng/l (n=13) was found for this matrix with a range <2500 to 84 000 ng/l. Additionally, in apple juice TFA was detected, but at lower levels (mean 6 200 ng/l; n=9). Significant levels of TFA (>25 000 ng/l) were found in two of the nine purees analysed. TFMS was frequently observed in orange juice as well (mean 16 ng/l; n=20), but only occasionally in apple juice. Both conventional and organic product were analysed. Generally, the organic samples showed lower or non-detectable TFA levels with two notable exceptions. Concentration of TFA was compared to drinking water guideline values from The Netherlands (NL), Denmark (DK) and Germany (DE). All samples with measurable TFA concentrations exceeded the NL value, frequently surpassed the DK limit, and one juice passed the DE limit. It was not within the scope of this study to assess origins of the products, sources for TFA/TFMS or to perform any risk assessment, although some general observations are being discussed.

Abbreviations:

DE = Germany; HFO-1234yf = 2,3,3,3-tetrafluoropropene; LIB = Lithium ion battery; LOQ = Limit of quantification; MRL = Maximum residue level; NL = The Netherlands; NO = Norway; ORU = Örebro University (SE); PBT = Persistent, bioaccumulative, toxic; PFAS = Poly- and perfluoroalkyl substances; PFAS4 = sum of PFOS, PFOA, PFHxS, PFNA; RPF = Relative potency factor; SE = Sweden; SPE = Solid phase extraction; TFA = trifluoroacetic acid; TFMS = trifluoromethane sulphonic acid; TDI = Tolerable daily intake; TWI = Tolerable weekly intake; UPLC-MS/MS = Ultra performance liquid chromatography – mass spectrometry/ mass spectrometry; vPvB = Very persistent, very bioaccumulative;

Introduction

Per- and polyfluoroalkyl substances (PFAS) have been high on the agenda concerning environmental problems over the past decade. Both environmental contamination and human exposure of PFAS have received considerable attention. PFAS have been manufactured and used since the 1950's, for example, in fluoropolymers, surface coatings, firefighting foams, cosmetics, paints, etc.

According to the most recent PFAS definition published in an OECD report, substances containing at least one fully fluorinated methyl or methylene carbon is to be classified as a PFAS (OECD, 2021). This has broadened the scope and more substances have been included. Ultrashort PFAS are characterized as having one, two or three carbon atoms. Among the ultrashort PFAS, two compounds have drawn the most attention, trifluoroacetic acid (TFA) and trifluoromethane sulphonic acid (TFMS; triflic acid). A growing number of reports show the presence of ultrashort PFAS in the environment, especially in water, but also plants, animals and foodstuffs (Neuwald et al., 2022; Garavagno et al, 2024). They are all associated with high water solubility and low pKa (acid dissociation constant) values, suggesting that, in the long-term the aquatic environment becomes the eventual sink. Therefore, they have been highlighted as potential vPvM and possibly PMT substances (UBA, 2023).

TFA can have a number of sources. One important source in precipitation is phototrans-formation of “modern” refrigerants such as HFO-1234yf, among others. (UBA, 2021). Other sources that have been suggested include the combustion of fluoropolymers, oxidative treatment of PFAS at remediation, direct use as an industrial chemical, and effluents from landfills and treatment plants (UBA 2021; Freeling and Björnsdotter, 2023; Garavagno et al., 2024).

TFA may also be formed at degradation of pesticides containing a CF_3 group (e.g. fluazinam, fluopyram) and at present there are around 40 such pesticides approved in the EU (UBA, 2021) and about 25 in use in the UK (Fidra, 2024) . Theoretical calculations by UBA (2021) indicate that decomposition of pesticides could be a major source. TFA and CF_3 moieties thus form a link to the concept of “PFAS pesticides” (PAN, 2023; 2024). These pesticides are classified as PFAS according to the recent definition and several of them can be found in fruit and vegetables. The PAN (2024) report showed that in 2021 “PFAS pesticides” were found in 20% of EU-grown fruit/berries and 12% of vegetables. The figures for imports to the EU were 18% for fruit and 12% for vegetables. However, MRL (max residue levels) values were not exceeded. It should be noted, though, that any residue of “PFAS pesticides” indicates their use but not the total potential of TFA formation.

There is a growing body of evidence that plants absorb and accumulate TFA. Uptake may occur through the root-water-soil system or via direct atmospheric uptake in leaves (Garavagno et al., 2024 and references therein). In some plants (bio)accumulation can be significant with factors ranging from 5-1 400 (Garavagno et al., 2024). Furthermore, trend data for European tree species show that TFA levels increased substantially between 1989-2020 (2-5 times; range 25-1060 µg/kg; Freeling et al, 2022). Regarding degradation of CF_3- containing pesticides, Chang An et al. (2017) demonstrated in experiments with wheat, lettuce, radish, carrot and spinach, that >10% of the ^{14}C signal from labelled pesticides added to the soil ended up as ^{14}C-TFA in the plants.

TFMS is a “super acid” used in organic synthesis and can be included as an additive in lithium ion battery (LIB) electrolytes (Neuwald et al., 2022). Less is known about TFMS compared to TFA, but Björnsdotter (2021) found TFMS (0.2-3 ng/l) in snow from Spitsbergen, Svalbard (NO). TFMS is also observed in drinking water as reported by van Hees et al. (2023; <2 ng/l) for Sweden and Norway and Neuwald et al. (2022; median 8 ng/l) for Germany. Regarding pesticides, there is limited information, but at the very least Fipronil contains a CF_3SO group alongside a CF_3 group. The substance is not allowed to be used in agriculture in the EU.

Findings on TFA distribution in the environment, together with data for pesticide use and TFA accumulation in plants, indicate that TFA enter the food chain and that human exposure occurs. Fruits and vegetables may thus, together with drinking water, be a significant exposure route.

The objective of this study was to investigate the presence of TFA and TFMS in juice and juice-based drinks. This included juice/drinks from various fruits, as well as puree-based products, particularly those aimed at babies and young children.

Material and Methods

A range of fruit and vegetable-based juices, drinks and purees (n=46) were purchased from local supermarkets in Lidköping (SE) over the period Feb-July 2024. See figures for further information. Products were stored refrigerated and/or frozen until analysis. As a reference, a number of oranges (n=7) were squeezed by hand, and the juice analyzed.

Analysis of ultrashort PFAS was performed by a method developed at Eurofins SE. The method is based on mixed mode (reversed phase/anion-exchange) separation using UPLC-MS/MS. The limit of quantification (LOQ) varied between typically 1-5 ng/l (TFMS) and 2 500-5 000 ng/l (TFA) in this investigation. A sub-set of samples (n=20) was analysed at Örebro University (ORU) where a supercritical fluid chromatography (SFC) system coupled with MS/MS was employed. The samples were subjected to SPE pre-concentration and LOQ was 500 ng/l for TFA and 20 ng/l for TFMS. Further details on the method can be found in e.g. Björnsdotter et al. (2022).

Results and Discussion

TFA and TFMS concentrations in juices and purees

Altogether 37 juices and drinks were analyzed as well as nine purees aimed for infants. (Fig 1-2). With a few exceptions, significant levels of TFA were found in the products. The highest concentrations were observed for orange juice and significant levels were found in 13 out of 21 samples. The average was 34 000±20 000 ng/l (n=13; mean ± S.D.), based on the assumption that all concentrated products were diluted five times (as recommended) with water free from TFA, and samples <LOQ were excluded. A corresponding number for apple juice would be 6 200 ±2 000 ng/l (n=9) if juices with no detectable TFA were omitted. As expected, the drinks with partial juice content (12% in ready-made product when stated) showed lower values (<LOQ to 740 ng/l). For the apple drink with a juice content of 12%, an estimation assuming that the juice was the only source of TFA yielded 6 200 ng/l, which is the same as the average value. The tomato juice showed a value between that of apple and orange juice, as did the mixed triple-fruit juice. For the latter, a calculation based on the mean levels in apple and orange juice (not considering the passionfruit) gave a concentration of 19 000 ng/l, close to the TFA content determined (17 000 ng/l).

Regarding TFA levels in the purees (Fig 2), significant levels were found in two cases, likely reflecting the purees and juices used. The products had different compositions and since no analysis of pure ingredients was performed no conclusions of relative contributions from the different fruits and vegetables could be made.

The TFA concentrations in this study can be compared to the values compiled in the review by Garavagno et al (2024). TFA was found in beer (median 6 100 ng/l; max 51 000 ng/l) and tea

(2400 ng/l). Levels in grains were comparable or even higher (16 000-180 000 ng/kg). Generally, much higher TFA concentrations than can be expected in drinking water were observed for the juice samples (see below).

Additionally, TFMS was identified, particularly in orange juice (16±23 ng/l; mean ± S.D; n=20). The maximum level determined was 110 ng/l, which was for a hand-squeezed orange and coincided with the highest TFA value recorded overall (84 000 ng/l). TFMS was found in both organic and conventional samples, but with a slight tendency for lower values in the former. In apple juices and drinks, TFMS was not identified with one exception while a value comparable to orange was obtained for the tomato juice. Concerning purees, TFMS is still being investigated and results will be presented later on. To the best of our knowledge, this is the first report on this ultrashort PFAS in drinks for consumption.

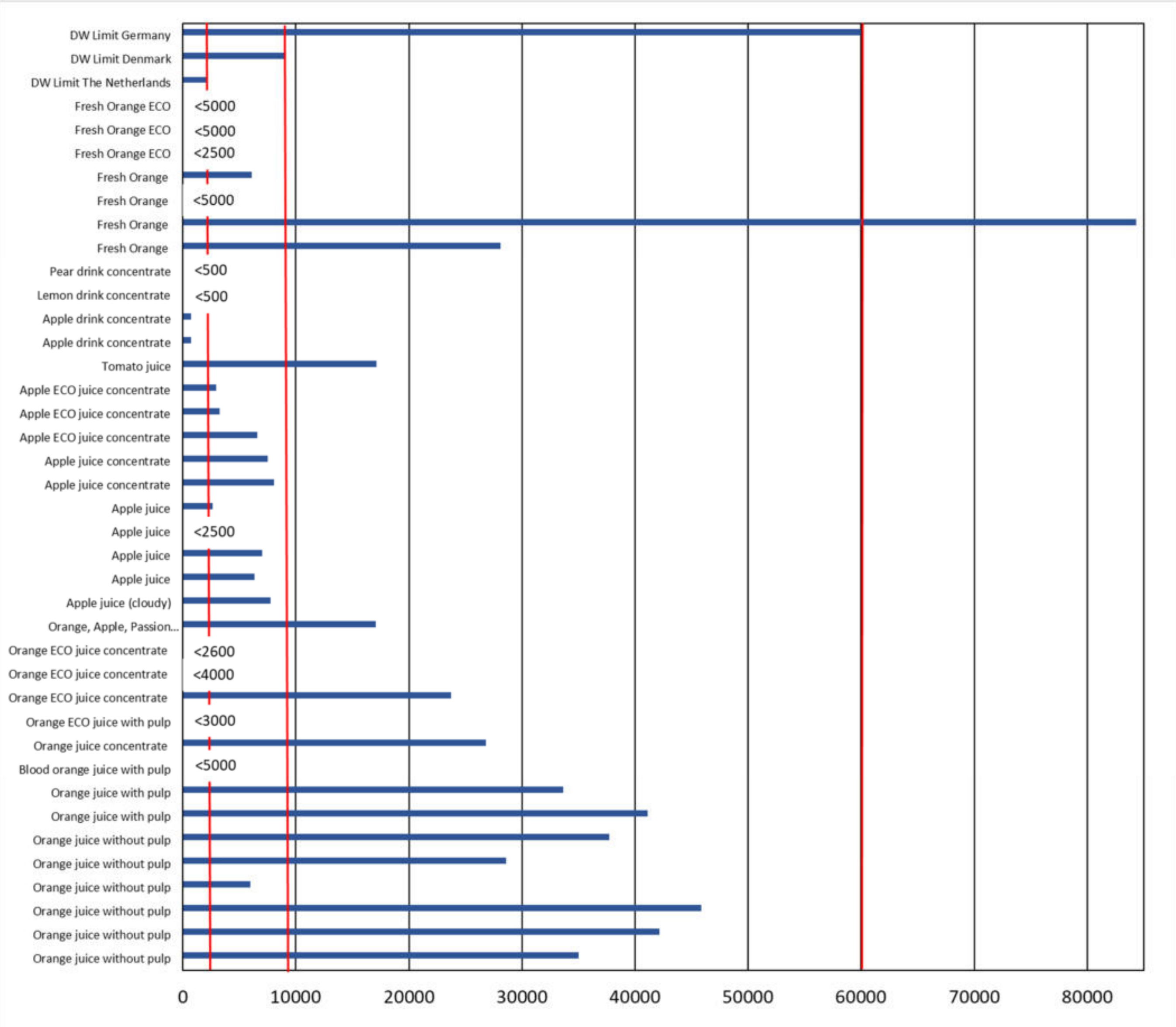

Fig. 1. TFA concentrations (ng/l) in juices and drinks. "ECO" denotes organic products and "Fresh" hand-squeezed orange juice. Values for concentrates refer to diluted products (1:5). As a reference drinking water (DW) guidelines for NL, DK and DE are included (red vertical lines)

To compare the values obtained by Eurofins F&F, both qualitatively and quantitatively, 20 samples were also analyzed at Örebro University (ORU). Good agreement (R^2=0.98; n=9) between analyses was observed for the orange juices despite different chromatographic approaches, and ORU TFA values corresponded to 118±11% of the Eurofins ones. For the apple-based products, a larger difference was found, with ORU measurements being approximately a factor 1.5 higher. The reasons for this discrepancy are currently being investigated. Nevertheless, a good correlation was observed (R^2=0.97; n=11). Likewise, for TFMS, the ORU analysis confirmed the order of magnitude, but for the four samples where a direct comparison could be made somewhat higher values were found at ORU.

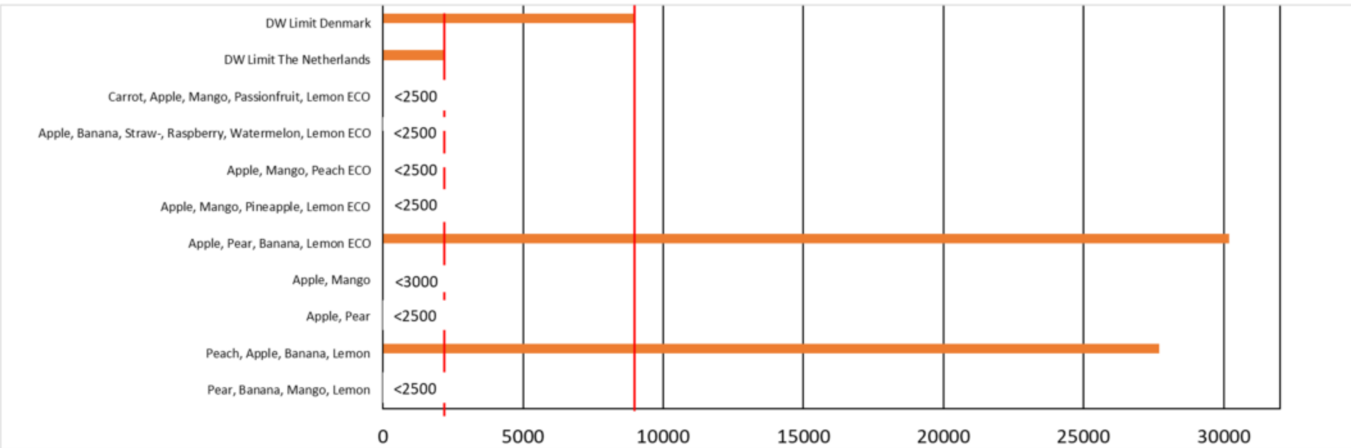

Fig. 2. TFA concentrations (ng/l) in purees. "ECO" denotes organic products. As reference drinking water (DW) guidelines for NL and DK are included (red vertical lines)

Although not part of the current work, 35 "regular" PFAS compounds (≥C4) e.g. PFOS and PFOA were tested for seven orange and apple juices (LOQ 10-20 ng/l). No detectable concentrations were observed.

Differences between products and origin

For the orange juice, it can be noted that the difference in TFA among conventional fresh or concentrated juice, with or without pulp, was within a factor of two for most part of the data set. Organic orange juice, including hand-squeezed one, was below the TFA reporting limit, with one exception. This "odd" sample showed a level corresponding to the conventional ones. The hand-squeezed oranges displayed a wide range of values. Regarding the apple juice, TFA could be found in both organic and non-organic produce, often at comparable concentrations. Regarding purees, it is not clear why two samples had significantly higher TFA levels than the others, which warrants further investigation. It should be noted that one of the two was labelled as organic.

The country of origin for the fruit used in production of juices was not always stated. In the current investigation, orange juice from Brazil, Italy and Spain/Greece were provided, while for apple juice, Poland and Sweden were mentioned. Otherwise, EU or non-EU origin was frequently stated. The finding that organic produce in many instances showed levels of both TFA and TFMS, even comparable to conventional ones for some, is of interest and more information about the sources are needed. Moreover, for apple juice, the two samples with the lowest TFA were both made from Swedish apples and were not labelled as organic. Regarding country of origin for orange juice, along with the distinction between juice vs hand squeezed ditto, the assessment becomes complex. For the four products originating from Brazil, the TFA levels ranged from 34 000 to 46 000 ng/l (all ready-made). The samples originating from Italy and Spain/Greece on the other hand, amounted to 6 000 ng/l TFA or were below the LOQ. No country specific information was given for conventional orange concentrate. Fresh oranges for squeezing were from Spain, with one exception from Egypt. The Spanish conventional oranges demonstrated the broadest range seen in the study <5 000 up to 84 000 ng/l. However, it can be anticipated that individual oranges may show a greater variation than batches of juice. The two highest levels were found for fruits of the "Lane late" variety. All organic oranges were also from Spain and all with non-detectable TFA levels. Regarding apple juice, besides the two Swedish samples with comparably low TFA concentrations (see above), the Polish sample had a value relatively close to the mean value calculated.

As described in the introduction, TFA can have several sources, and it has not been part of this study to assess or quantify contributions. However, an important aspect to consider is whether the water used to prepare "ready to drink" juices from concentrates can be a significant source of TFA. It is not always apparent on the labels if the juice/drink is made from concentrate or not. However, an overview of the samples where such comparisons can be made indicates that the water for dilution is unlikely to be a significant factor. Preparation (dilution) of orange juice has taken place in countries such as Sweden, Denmark, Germany and Spain. The Information on TFA levels in drinking water is not complete but existing data for Scandinavia (van Hees et al, 2023; unpubl) show a range between <50-720 ng/l and for Germany <LOQ-1 100 ng/l (Global 2000, 2024; van Hees et al., unpubl). The similar TFA concentration for both orange juices made from concentrate and fresh products, in relation to what can be expected in drinking water, strengthen this assumption. However, for "ready to drink" apple juices made from concentrates, process water could potentially be a contributing, though assumably not major, factor. Moreover, dilution of concentrates (juices and drinks) bought by consumers will likely add further TFA, but this is not considered in this work. This is the case also for any potential contribution from packaging material.

Comparison to guideline values

TFA guideline/limit values for drinking water exist in The Netherlands (NL), Denmark (DK) and Germany (DE) (UBA, 2020; RIVM, 2023a; Miljøministeriet, 2024). Among samples with TFA detected, all orange juice bought in stores, some hand-squeezed, as well as the tomato and "triple" juice, exceeded the NL limit of 2 200 ng/l. All apple juice (including organic) except one also passed this limit. The DK limit (9 000 ng/l) was exceeded repeatedly for orange juice as well. The highest TFA sample in the dataset (hand-squeezed; 84 000 ng/l) did not conform to the DE limit (60 000 ng/l) either. With regard to purees aimed for babies and small children, the two samples with measurable levels of TFA were both higher than the NL and DK guidelines. It can also be noted that a recent report from a group of European environmental NGOs (Global 2000, 2024) calculated alternative limits for TFA in drinking water, considering the intake of infants, ranging between 1 300-2 400 ng/l.

TFA may also be directly compared to a TDI or TWI (tolerable daily/weekly intake) value. In the method by RIVM (2023), TFA is included in a weighted sum of PFAS that is compared to EFSA's TWI value of 4.4 ng/kg bw/week for PFAS4. The weighting is based on relative potency factors (RPF) where TFA has a value of 0.002. Using this approach, a 10 kg child could drink 92 ml "average" orange juice (34 000 ng/l) per day to reach the limit (TWI ÷ 7). A similar volume, 105 ml per day would be obtained for the puree with the highest TFA level (approximately one pouch/day). It should be stressed that this calculation assumes no further PFAS exposure but

TFA. If the TDI values (1 000-1 800 ng/kg bw/day) derived for TFA alone in the Global 2000 (2024) study were used, this would imply a daily consumption of 290-530 ml of average orange juice or 3-6 “worst case” 100 ml puree poaches. In contrast, if the TDI underpinning the current German limit value was used, a 10 kg child could ingest up to 5 300 ml of orange juice per day. Similar calculations for adults would yield higher tolerable volumes.

Conclusions

This work gives, to our knowledge, the first overview of ultrashort PFAS, TFA and TFMS, in juice, juice-based drinks and fruit purees for children. The concentrations of TFA, particularly in orange juice, appear high when compared to drinking water and other fruits tested, but are regarded as realistic given the increased knowledge of TFA contamination and uptake into plants. TFA and TFMS were also detected at a significant level in some samples of organic produce. It has not been within the scope of this work to determine the origin of TFA and processes of uptake, but there are probably multiple sources that could potentially contribute. Nor was it the intention to draw conclusions on exposure and toxicity; however, the levels found are still relevant compared to (or higher than) guideline limits for drinking water. Intake calculations for children (bodyweight = 10 kg) using current TDI estimates suggest that especially orange juice and some purees could result in a significant exposure. The finding of TFMS in juice is novel, and although concentrations are lower than for TFA, the substance still deserves to be assessed in the future.

Acknowledgements

The authors would like to thank Pontus Larsson (MSc; Örebro Univ) for conceptual ideas and discussion about the project and results as well as work with TFA and TFMS determinations.

References

Björnsdotter. M.K., 2021: Ultra-short-chain perfluoroalkyl acids: Environmental occurrence, sources and distribution. Doctoral Thesis, Örebro Studies in Chemistry 29. Örebro University. https://oru.diva-portal.org/smash/get/diva2:1573193/FULLTEXT01.pdf

Björnsdotter, M. K., Yeung, L.W.Y., Kärrman, A., Ericson Jogsten, I. Mass Balance of Perfluoroalkyl Acids, Including Trifluoroacetic Acid, in a Freshwater Lake. Environ. Sci. Technol. 2022, 56, 251–

Chang An, K., Yng Choy, Y., Fleischmann, T., Dohn, D. Application note. Identification of trifluoroacetic acid as polar metabolite from pesticides containing a trifluoroacetic (CF_3) moiety using ^{14}C tracer technology. M-032717, EAG Laboratories, 2017

Fidra, 2024. PFAS active substances in UK pesticides. https://www.fidra.org.uk/download/pfas-in-uk-pesticides/

Freeling, F., Björnsdotter, M. K. Assessing the environmental occurrence of the anthropogenic contaminant trifluoroacetic acid (TFA). Curr. Opinion Green Sustain. Chem. 2023, 41, 100807

Freeling, F., Behringer, D., Heydel, F., Scheurer, M., Ternes, T. A., Nödler, K. Trifluoroacetate in Precipitation: Deriving a Benchmark Data Set. Environ. Sci. Technol. 2020, 54, 18-

Garavagno, M. D. L. A., Holland, R., Khan, M. D. H., Orr-Ewing, A. J., Shallcross, D. E. Trifluoroacetic Acid: Toxicity, Sources, Sinks and Future Prospects. Sustainability, 2024, 16, 2382-

Global 2000, 2024. TFA: The Forever Chemical in the Water We Drink. https://www.global2000.at/sites/global/files/TFAinWater_report_Final_EN.pdf

Miljøministeriet, 2024. Drikkevandsbekendtgørelsen. Bekendtgørelse om vandkvalitet og tilsyn med vandforsyningsanlæg, BEK nr 810 af 18/06/2024 (https://www.retsinformation.dk/eli/lta/2024/810)

Neuwald, I. J., Hübner, D., Wiegand, H. L., Valkov, V., Borchers, U., Nödler, K., Scheurer, M., Hale, S. E., Arp, H. P. H., Zahn, D. Ultra-Short-Chain PFASs in the Sources of German Drinking Water: Prevalent, Overlooked, Difficult to Remove, and Unregulated. Environ. Sci. Technol. 2022, 56, 10, 6380–

OECD, 2021. Reconciling Terminology of the Universe of Per- and Polyfluoroalkyl Substances: Recommendations and Practical Guidance, OECD Series on Risk Management, No. 61, OECD Publishing, Paris.

PAN (Pesticide Action Network), 2023. Europe's Toxic Harvest: Unmasking PFAS Pesticides Authorised in Europe. https://www.pan-europe.info/resources/reports/2023/11/europes-toxic-harvest-unmasking-pfas-pesticides-authorities-europe

PAN (Pesticide Action Network), 2024. Toxic Harvest: The rise of forever PFAS pesticides in fruit and vegetables in Europe. https://www.pan-europe.info/resources/reports/2024/02/toxic-harvest-rise-forever-pfas-pesticides-fruit-and-vegetables-europe

RIVM, 2023a. Risk assessment of exposure to PFAS through food and drinking water in the Netherlands. RIVM report 2023-0011, Bilthoven (https://www.rivm.nl/publicaties/risk-assessment-of-exposure-to-pfas-through-food-and-drinking-water-in-netherlands#:~:text=The%20study%20shows%20that%20the,be%20harmful%20to%20their%20health.)

RIVM 2023b; Advies Indicatieve drinkwaterrichtwaarde voor stof trifluorazijnzuur (TFA, CAS-nr. 76-05-1) https://www.rivm.nl/documenten/bijlage-bij-rivm-brief-aan-ilt-indicatieve-drinkwaterrichtwaarde-trifluorazijnzuur-tfa

UBA, 2020. Trifluoressigsäure (TFA)–Gewässerschutz im Spannungsfeld von toxikologischem Leitwert, Trinkwasserhygiene und Eintragsminimierung. Erläuterungen zur Einordnung des neuen Trinkwasserleitwerts von 60 µg/L. 20 Oktober 2020. Umwelt-bundesamt.https://www.umweltbundesamt.de/sites/default/files/medien/362/dokumente/2020_10_20_uba_einordnung_tfa_leitwert.pdf

UBA, 2021. Reducing the input of chemicals into waters: trifluoroacetate (TFA) as a persistent and mobile substance with many sources. ISSN 2363-829X. Umweltbundesamt, Dessau-Roßlau, DE

UBA, 2023. A prioritization framework for PMT/vPvM Substances under REACH for registrants, regulators, researchers and the water sector. TEXTE 22/2023. Umweltbundesamt, Dessau-Roßlau, DE

van Hees, P., Sundelin, T., Karlsson, P. 2023. Ultrashort PFAS in Swedish and Norwegian Drinking Water. https://www.eurofins.se/tjaenster/miljoe-och-vatten/nyheter-miljo/eurofins-study-ultrashort-pfas-in-swedish-and-norwegian-drinking-water/