DANISH ENVIRONMENTAL PROTECTION AGENCY

Potential sources of 1,2,4-triazole in Danish groundwater

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1 Dansk sammendrag

Der er i de seneste år blevet påvist stoffer i danske grundvandsprøver, som ikke indgår standardmæssigt i det nationale overvågningsprogram. Et af dem er 1,2,4-triazol, der er en metabolit af fungicider der anvendes i landbruget (kendt som azoler), men som også indgår i stoffer til andre formål, f.eks. træbeskyttelsemidler, nitrifikationshæmning, medicin mv.

Miljøstyrelsen har derfor ønsket en udredning af, hvilke forureningskilder der potentielt kan påvirke det danske grundvand med 1,2,4-triazol. Følgende kilder til forekomst af 1,2,4-triazol i miljøet er vurderet i dette notat:

- Nedbrydningsprodukt fra azol-fungicider og andre azolholdige pesticider anvendt i landbruget
- Nedbrydningsprodukt fra azol-fungicider anvendt som biocider i træbeskyttelsesmidler
- Nitrifikationshæmmer anvendt i landbruget
- Nedbrydningsprodukt fra azolholdige lægemidler
- Nedbrydningsprodukt fra azolholdige industrikemikalier
- Naturlig dannelse af 1,2,4-triazol i miljøet.

Nedbrydningsprodukt fra pesticider anvendt i landbruget
Der er identificeret 11 azol-fungicider, der anvendes eller har været anvendt i landbrugsafgrøder i Danmark, og hvoraf mellem 17% og 24% af molekylerne (på vægtbasis) udgøres af 1,2,4-triazol. Propiconazol er med en samlet mængde på omkring 1,8 millioner kg aktivstof over hele anvendelsesperioden klart det mest anvendte af disse fungicider, mens stoffer som tebuconazol og epoxiconazol hver har været brugt til landbrugsformål i en mængde på ca. 600.000 kg aktivstof.

Andre vigtige stoffer er (eller har været) bitertanol, prothioconazol, triadimefon og triadimenol, hvor det samlede salg af hver ligger i intervallet 174.000 til 463.000 kg aktivstof set over hele den periode, hvor de har været markedsført. Heraf udgør 1,2,4-triazol mellem 20 og 24%.

Desuden findes der et herbicid, amitrol, hvoraf 82% er 1,2,4-triazol, som blev solgt fra 1958-1989 i en samlet mængde på 443.000 kg aktivstof og som blev anvendt i høje doseringer; mellem 1,0 og 9,5 kg aktivstof/ha sammenlignet med 0,125-0,250 kg aktivstof/ha for azol-fungiciderne.

Et worst case estimat af potentialet for grundvandforurening med disse landbrugspesticider er, for hver udført sprøjtning, 11-30 gram/ha for fungiciderne og 1.950 gram/ha som gennemsnit for amitrol. Efterårssprøjtning med azolerne, som ikke længere er tilladt i Danmark (siden 2014), må anses for at være det mest kritiske anvendelsesscenarie i forhold til risiko for grundvandsforurening.

Nedbrydningsprodukt fra biocider i træbeskyttelsesmidler
Der er kun to azol-fungicider, der er relevante at vurdere i forhold til anvendelse som biocider i træbeskyttelsemidler i Danmark, nemlig propiconazol og tebuconazol. De er i perioden 2000-2017 blevet solgt til dette formål i samlede mængder på hhv. 172.000 og 167.000 kg aktivstof. De indgår i en række forskellige produkter, der spænder fra industrielle trykimprægneringsmidler til midler, der påføres træets overflade med pensel eller lignende. Koncentrationen af azol-fungicid i overfladelaget vurderes typisk at være i størrelsesordenen 0,006-0,007 kg aktivstof/m² (hvoraf 20-22% er 1,2,4-triazol).

Hvor landbrugsfungiciderne giver anledning til en jævn belastning over store arealer er belastningen fra anvendelsen til træbeskyttelse koncentreret lige omkring de steder, hvor det imprægnerede træ produceres, oplagres eller anvendes til huse, stakitter og andre trækonstruktioner.

Nitrifikationshæmmer i landbruget
1,2,4-triazol indgår i visse landbrugsgødninger som nitrifikationshæmmer, men det vurderes, at omfanget af denne anvendelse på nationalt plan i Danmark er meget begrænset, og at den derfor har meget lille betydning på det overordnede plan, men eventuelt kan bidrage lokalt på de specifikke arealer. Der blev for få år siden solgt mindre mængder af denne type nitrifikationshæmmer, men der er ikke fundet oplysninger, der kan bekræfte, at nitrifikationshæmmere med 1,2,4-triazol stadig anvendes.

Nedbrydningsprodukt fra azolholdige lægemidler
Der er identificeret i alt 11 lægemidler i Danmark, der indeholder en 1,2,4-triazolgruppe, som vil kunne frigives ved nedbrydning af aktivstoffet og primært vil afgives til spildevand. Det mængdemæssigt dominerende af de 11 aktivstoffer er fluconazol, som estimeres at repræsentere omkring 90% af det samlede 1,2,4-triazolpotentiale fra danske lægemidler. Det er estimeret som worst case, at 17 kg 1,2,4-triazol/år hidrørende fra fluconazol kan blive tilført landbrugsjord gennem udbringning af spildevandsslam.

Nedbrydningsprodukt fra industrielle anvendelser
Ifølge REACH registreringsdossieret for 1,2,4-triazol anvendes stoffet som intermediær ved fremstillingen af agrokemikalier, lægemidler og som nitrifikationshæmmer. I den nordiske SPIN-database, som sammenfatter data fra de nordiske produktregistre, er der registreret 56 stoffer, som indeholder "1,2,4-triazol" i det kemiske navn. Data for disse 56 stoffer er blevet indhentet fra det danske Produktregister samt fra SPIN-databasen, som indeholder de ikke-fortrolige oplysninger.

De vigtigste ikke-landbrugsmæssige anvendelser er i træbeskyttelsemidler, som beskrevet separat ovenfor, mens i alt 0,4 tons/år blev brugt til forskellige typer af maling, sandsynligvis udendørs malinger og træolier med afgivelsesmønster til miljøet svarende til træbeskyttelsesmidlerne. Derudover er ca. 0,1 tons/år benyttet i smøremidler og hydralikvæsker, hvor stoffets funktion er at beskytte mod korrosion. Miljøpåvirkningen fra denne anvendelse anses for ubetydelig.

Naturlig dannelse af 1,2,4-triazol i miljøet

Det er blevet påvist, at 1,2,4-triazol kan dannes naturligt af nogle grupper af mikroorganismer i jordmiljøet. Datagrundlaget er ret spinkelt, men resultaterne af de udførte undersøgelser indikerer, at den beregnede årlige dannelse af 1,2,4-triazol i skovjorde per hektar kan være af samme størrelsesorden som den mængde, der kan frigives ved en sprøjtning med azol-fungicider i landbruget. Forskellen på påvirkningen ift. grundvand er især, at i landbruget bringes hele mængden ud på én gang, mens den naturligt dannede 1,2,4-triazol frigives jævnt over året og derfor lettere kan blive nedbrudt inden den siver ned under de øverste jordlag. Det vides i øvrigt ikke, om den naturlige dannelse er lige så stor i landbrugsjorde som i mere humusrige jorde i skove.

Sammenfatning og samlet vurdering

Baseret på den information, der har været til rådighed for opgaven, vurderes det overordnet, at det er den landbrugsmæsige anvendelse af azolholdige pesticider, særligt azol-fungicider, der repræsenterer det største potentiale for udvaskning af 1,2,4-triazol til grundvandet i Danmark. Det er frem for alt ved sprøjtning om efteråret, at der er risiko for udvaskning, idet der i den periode normalt både er overskud af nedbør og ret lave temperaturer, der nedsætter hastigheden for mikrobiel nedbrydning af stoffet. Det skal dog nævnes, at efterårsudbringning af azol-fungicider ikke har været tilladt i Danmark siden 2014.

Amitrol har i sin tid kunnet udgøre en betydelig forureningskilde, der hvor stoffet er benyttet, dels i kraft af den meget høje dosering og det høje indhold af 1,2,4-triazol (82%), dels fordi den største anvendelse var til bekæmpelse af ukrudt i stubmarker om efteråret. Umiddelbart skulle man dog forvente, at 1,2,4-triazol hidrørende fra amitrol var helt nedbrudt på nuværende tidspunkt, 30 år efter, at salg af dette ukrudtsmiddel ophørte i Danmark.

Lokalt kan anvendelse af azol-fungicider i træbeskyttelsesmidler og som nitrifikationsinhibitor muligvis have en vis betydning, mens andre produkter og anvendelser vurderes at være af ingen eller meget ringe betydning.

Det kan ikke afvises, at naturlig dannelse af 1,2,4-triazol i jordmiljøet kan være medvirkende til stoffets forekomst i grundvand.

2 Introduction

Danish groundwater and drinking water has for many years been monitored regularly for its content of a substantial number of pesticides and pesticide metabolites with the aim to reveal possible contamination due to plant protection activities in agriculture, horticulture and forestry, as well as on non-cultivated land.

However, the substance 1,2,4-triazole, which is a component and possible metabolite of a number of widely used fungicides (azole fungicides), is not part of the standard pesticide monitoring programme in Denmark but has recently been detected in samples of groundwater and drinking water undergoing extended investigations.

This observation has led to considerable public debate and political concern. In response to this, the Danish Environmental Protection Agency (DEPA) has decided to initiate this survey aiming to provide an overview of possible sources of 1,2,4-triazole in Danish groundwater, also from non-pesticidal and non-biocidal uses and including possible natural formation of the substance.

3 Scope and methodology

According to the scope of work for this survey requested by the DEPA, the following known or possible sources of 1,2,4-triazole in relation to groundwater contamination should be included:

- Degradation product from azole pesticides, primarily fungicides, used as agricultural plant protection products
- Degradation product from azole fungicides used as biocides for preservation of wood
- Nitrification inhibitor used in agriculture
- Degradation product from medicinal products used to control fungal diseases and various other pharmaceutical uses
- Degradation product from industrial chemicals containing a 1,2,4-triazole moiety. This also includes biocidal uses other than preservation of wood.
- Natural formation of 1,2,4-triazole
- Possible other realistic sources.

Agricultural pesticides:

The description and assessment of agricultural pesticides as a contamination source for 1,2,4-triazole in groundwater is based on pesticide sales data obtained from the Danish EPA's pesticide database, other information on the uses of the relevant active substances for plant protection purposes in Danish agriculture (e.g. "Middeldatabasen" and "Vejledning i planteværn" (SEGES)), and on approved EU endpoints for environmental fate (degradation rates, formation of metabolites etc.) of selected active substances as reflected in the EFSA conclusions. The latter information is used for calculation of the "1,2,4-triazole potential" for selected substances.

Wood preservation fungicides:

As for the azole pesticides, the main source of information on biocidal uses of azole fungicides for preservation of wood is the Danish EPA's pesticide database. The reason for this is that, historically, active substances and products within this use category have been regulated by the Danish EPA as part of the national legislation and administration of pesticides.

With regard to the possible environmental exposure resulting from the use of azole substances as wood preservation agents, information on specific approved uses of relevant products has been obtained from the Danish EPA's "Oversigt over godkendte bekæmpelsesmidler" (List of approved pesticides), which has revealed products for industrial preservation of wood, including pressure impregnation, as well as for non-professional/domestic application by brush.

Nitrification inhibitor:

1,2,4-triazole is used as nitrification inhibitor in two types of products: liquid nitrification inhibitor added to organic fertilisers (manure) and solid fertilisers with nitrification inhibitor as additive. For the liquid nitrification inhibitors information has been obtained from the literature and contact to suppliers of liquid nitrification inhibitors in Denmark. For the solid fertilisers it has been investigated if fertilisers with nitrification inhibitors listed in the Danish fertiliser inventory (in Danish: "Gødningsfortegnelse fra Landbrugsstyrelsen") contain 1,2,4-triazole. It is mandatory to indicate in the inventory if the fertilisers contain nitrification inhibitors. It has furthermore been investigated whether fertilisers known to contain 1,2,4-triazole are included in the list.

Medicinal products:

A list of medicinal products that may contain a 1,2,4-triazole moiety has been generated on the basis of information from the literature and the Danish medicinal products database pro.medicin. For the identified substances, data on the consumption in Denmark have been extracted from the Danish Register of Medicinal Products Statistics (in Danish: "Lægemiddelstatistikregistret") of the Danish Medicines Agency. The consumption is in the database quantified as defined daily doses (DDD). The DDD is a standard daily dose established by the WHO for each medicinal product and route of administration and information on DDD has been retrieved from WHO ATC/DDD Index 2018.

Information on pharmacokinetics, i.e. information on the fate of the medicinal product in the body, has been retrieved from the Danish medicinal products database pro.medicin. Information on the fate of the products during waste water treatment has been obtained from the scientific literature.

Industrial chemicals:

Information on the possible use of industrial chemicals containing a 1,2,4-triazole moiety has initially been retrieved from the REACH registration dossier for 1,2,4-triazole and the websites of the registrants. Furthermore, a search for chemicals with the string “1,2,4-triazole” in the chemical name in the REACH registration database showed that 92 substances with the text string “1,2,4-triazol” in the chemical name are registered under REACH. Many of the registered substances are used as intermediate only.

In order to investigate which of these may be used in Denmark, a list of substances with the text string “1,2,4-triazol” was retrieved from the SPIN database of the Nordic product registers. The list contained in total 56 substances and data for these has been retrieved from the Danish Product Register, December 2018 and from the SPIN database (latest year, 2016). The SPIN database includes non-confidental information from the Danish Product Register and these data was used for the further assessment. For the main identified uses further information has been obtained from an internet search of Safety Data Sheets and manufacturer’s websites.

Natural formation in the environment:

Information on the possible natural formation of 1,2,4-triazole in the environment has been obtained from a number of recent conference proceedings (non-peer reviewed papers) and from other literature. Searches has been done with relevant search strings in the bibliographic database pub.med.

Other possible sources:

Information on other possible sources has been retrieved through broad internet searches and searches in bibliographic databases using relevant search strings. Information on used volumes for ammunition has been retrieved from the REACH registration database at ECHA’s website.

4 Agricultural 1,2,4-triazole pesticides

4.1 Consumption and uses of azole pesticides

4.1.1 Azole fungicides

A number of azole pesticides are or have previously been used in Danish agriculture, primarily fungicides for control of a variety of fungal diseases in important field crops such as cereals, oilseed rape, seed crops and beets. The relevant active substances (a.s.) are listed in Table 4-1 below together with their periods of use/consumption and amounts of a.s. sold.

Table 4-1 Overview of agricultural 1,2,4-triazole-based fungicides and their consumption in Danish agriculture.

Active substance	Time period of sale in DK	Total consumption (kg a.s.)	Min.- max. consumption (kg a.s./year)
Bitertanol	1987 - 2011	398,939	731 – 38,099
Difenoconazole	1998 - present	53,912	577 – 4,466

Active substance	Time period of sale in DK	Total consumption (kg a.s.)	Min.- max. consumption (kg a.s./year)
Epoxiconazole	2003 - present	607,442	13,259 – 63,349
Metconazole	2003 - present	14,740	0 – 4,464
Paclobutrazole**	1990 - present	648	3 – 52
Penconazole**	1995	124	124
Propiconazole*	1982 - present	1,980,763	9,045 – 184,797
Prothioconazole	2006 - present	463,100	7,395 – 104,437
Tebuconazole*	1996 - present	798,142	300 – 79,433
Triadimefon	1977 – 1993	249,833	42 – 59,121
Triadimenol	1981 - 1995	174,216	141 – 41,836

* Total consumption for use as agricultural fungicide and biocide for wood impregnation

** Only for indoor use in greenhouses.

The trends over time in the agricultural use of the most important azole fungicides used in Danish agriculture is shown in Figure 4-1 (consumption) and Figure 4-2 (distribution on crop types) (Miljø- og Fødevareudvalget 2016-17. MOF Alm. del endeligt svar på spørgsmål 1017. Prepared by Aarhus University, Institute of Agroecology, 21 September 2017).

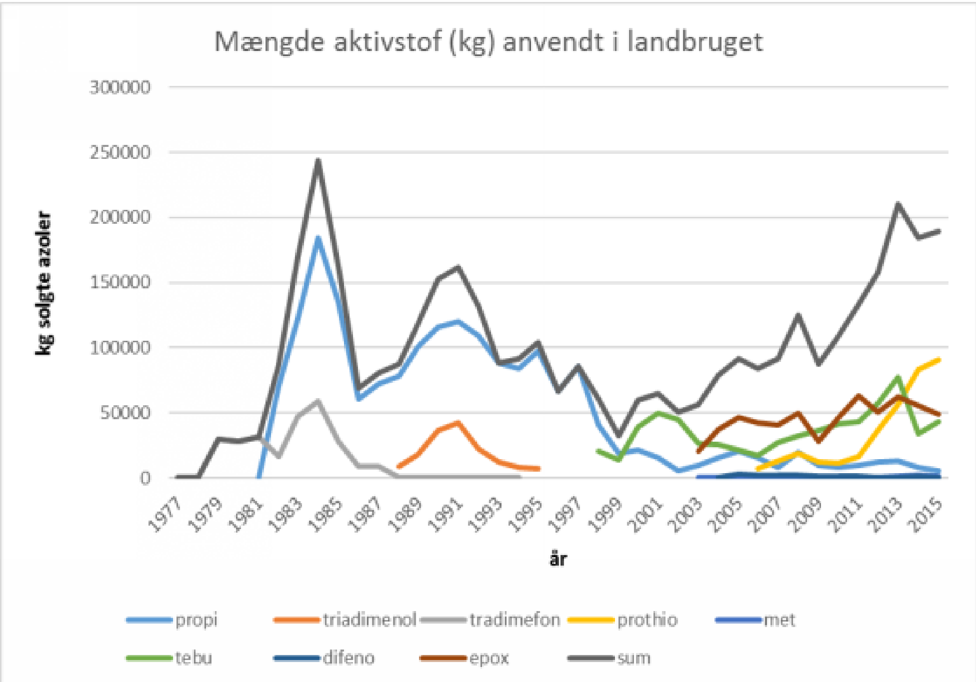

Figure 4-1 Trends in the consumption of agricultural azole fungicides in Denmark from 1977 to 2015 (Source: MOF Alm. del endeligt svar på spørgsmål 1017, offentlig, 2017).

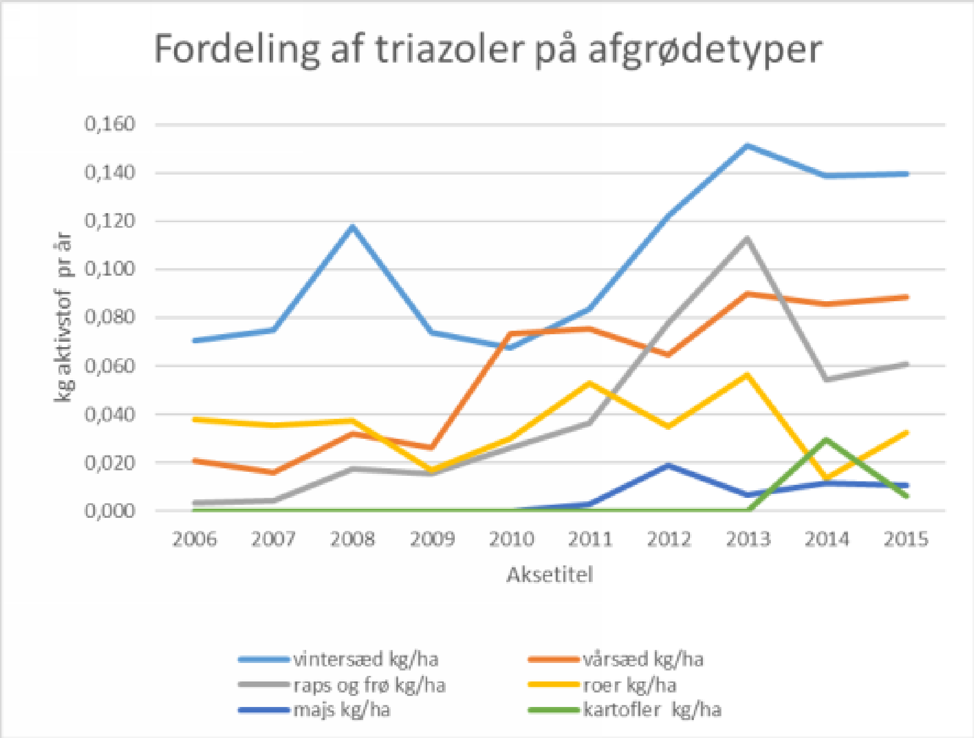

Figure 4-2 *Use of agricultural azole fungicides in Denmark, 2006-2015, distributed on field crops (Source: MOF Alm. del endeligt svar på spørgsmål 1017, offentlig, 2017).*

Thus, by far the largest amounts of the azole fungicides are being or have been used in large field crops in Denmark but two of the substances, propiconazole and tebuconazole, are also being used for preservation of wood against deterioration caused by attack by fungi (biocidal use, see section 5).

Some of the substances have not only been applied on field crops by spraying but also been used for seed treatment, however at much lower dosages per hectare than when used to control leaf fungal diseases. The active substances used for seed treatment are bitertanol, difenoconazole, propiconazole, tebuconazole and triadimenol. Typical dosages for field spraying are 125-250 gram/hectare against 1.5-56 gram/hectare for treatment of seeds. In terms of total tonnage, the agricultural field uses of the pesticides are the most important (see section 5) and only minor amounts are used for seed treatment purposes.

4.1.2 Non-fungicidal azole pesticides

One non-fungicidal azole pesticide, namely the herbicide amitrole, has been used extensively in Danish agriculture[1]. Amitrole was used from 1958-1989 in a total amount of almost 443,000 kg a.s., see Table 4-2.

[1] The herbicide florasulam also contains a 1,2,4-triazole moiety. However, according to the EFSA conclusion for this herbicide, 1,2,4-triazole is not formed as a metabolite during its degradation in soil.

Amitrole was applied at much higher rates than the azole fungicides, between 1 and 9.5 kg a.s. per hectare. The substance was mainly used post-harvest in field crops (only approved on stubble fields) but also for total weed control in hedgerows, on uncultivated areas, under fruit trees and in forests.

Table 4-2 Consumption of the azole herbicide amitrole in Danish agriculture.

Active substance	**Time period of sale in DK**	**Total consumption (kg a.s.)**	**Min.- max. consumption (kg a.s./year)**
Amitrole (herbicide)	1958 - 1989	442,937	506 – 34,695

A couple of insecticides, triazamate and triazophos, also contain 1,2,4-triazole. However, only triazophos has been registered for use in Denmark (from 1979 to 1990). Triazophos was presumably only used in small amounts and mainly in the summer period and is therefore considered negligible in the current context.

4.2 Environmental exposure from use of azole pesticides

4.2.1 1,2,4-triazole potential

The use of azole substances as plant protection agents in agricultural crops lead in principle to close to full (100%) environmental exposure as the substances are applied on the fields and only small amounts are disposed of elsewhere as waste. However, the 1,2,4-triazole moiety only constitutes a fraction of the pesticide molecule and, hence, the "1,2,4-triazole potential" of the active azole substances is, in most cases, considerably less than the a.s. amounts appearing in the annual pesticide sale statistics.

An overview of the 1,2,4-triazole potential of the main azole pesticides is presented in Table 4-3 (based on their total sales in Denmark over the years).

Table 4-3 1,2,4-triazole potential of azole pesticides used in Danish agriculture.

Active substance	**Total consumption (kg a.s.)**	**Hereof 1,2,4-triazole (%)**	**Total 1,2,4-triazole potential (kg a.s.)**	**1,2,4,-triazole potential, "field rate" (kg/ha)****
Amitrole	442,937	82	363,208	3.90
Bitertanol	398,939	20	79,820	0.010
Difenoconazole	53,912	17	9,165	0.021
Epoxiconazole	607,442	21	127,563	0.026
Propiconazole	1,980,763*	20	396,153	0.025
Prothioconazole	463,100	20	92,620	0.040

Active substance	Total consumption (kg a.s.)	Hereof 1,2,4-triazole (%)	Total 1,2,4-triazole potential (kg a.s.)	1,2,4,-triazole potential, "field rate" (kg/ha)**
Tebuconazole	798,142*	22	175,591	0.055
Triadimefon	249,833	24	59,960	0.060
Triadimenol	174,216	23	40,070	0.058
Total	**5,169,284**	**-**	**1,344,150**	**-**

* This figure includes the amount used for preservation of wood (biocidal use) up to and including 2016.

** Based on standard dosages for field spraying. Amitrole based on a dose rate of 4.75 kg a.s./ha (50% of max.). Bitertanol has only been used for seed treatment.

More detailed information on the sales of propiconazole and tebuconazole for agricultural purposes indicates that for the period 2000-2017 about 50% of the amount of propiconazole (roughly 339,000 tons corresponding to 17% of the total sale (starting 1982)) and 70% of the amount of tebuconazole (roughly 828,500 tons corresponding to 91% of the total sale (starting in 1996)) were applied by field spraying in agricultural crops. See Table 4-4 for more details.

Table 4-4 Consumption of propiconazole and tebuconazole used as agricultural fungicides from 2000-2017 as compared to the total consumption.

Period	Total kg a.s.	Pesticide kg a.s.	Pesticide % of total
Propiconazole			
2000-2017	339,165	171,955	51
2007-2017	166,241	102,385	62
2000-2006	172,924	69,570	40
Tebuconazole			
2000-2017	828,588	572,946	69
2007-2017	564,708	538,746	95
2000-2006	263,880	34,200	13

A significant shift in the use pattern of propiconazole and tebuconazole seem to have occurred around 2007, most dramatically for tebuconazole, which until and including 2006 was primarily used as a wood preservative but since 2007 it has almost exclusively been used as an agricultural fungicide (based on total sales data for 2000-2017 and detailed sales data for agricultural field uses for the same period). The change for propiconazole is less pronounced but the vast majority of the total sale of this active substance, more than 80%, took place before 2000. During that period, it is beyond doubt the pesticidal uses in agriculture that are completely dominant although specific figures on the distribution have not been available for the preparation of this note.

Therefore, the agricultural part of the total "1,2,4-triazole potential" in Table 4-3 is estimated to be around 350,000 tons for propiconazole (for the whole period, 1982-2017) and around 140,000 tons for tebuconazole. For the other azole pesticides, the 1,2,4-triazole potential remains unchanged, i.e. 100% for agricultural uses and other applications in forestry and on uncultivated land.

4.2.2 Formation of 1,2,4-triazole from azole pesticides

Information on the formation of 1,2,4-triazole and other metabolites during degradation of some of the main agricultural fungicides (difenoconazole, epoxiconazole, propiconazole and tebuconazole) and the herbicide amitrole was obtained from the respective EFSA conclusions (including the lists of endpoints; LoEP).

There is some difference in the aerobic soil degradation rate studies with regard to the formation of 1,2,4-triazole as a metabolite, i.e. whether the substance is formed in amounts higher or lower than 10% AR (the Application Rate) during degradation in soil. However, making an average consideration, the approximate maximum observed level of 1,2,4-trizole formation appears to be about 10% AR.

From the EFSA conclusions and from the EU Commission's proposed revision to DT_{50} (time for 50% dissipation of a substance) of 1,2,4-triazole (EU Commission, 2013), it can be deduced that the aerobic soil degradation of 1,2,4-triazole has a biphasic profile where the degradation takes place rapidly during the first week of incubation and thereafter at a much slower rate. During the first approximately one week of incubation in studies with three soils using 1,2,4-triazole as parent compound (EU Commission, 2013), the concentration of untransformed 1,2,4-triazole was reduced to about 30-45% of the applied amount. After 120 days (at termination of study), the concentration of 1,2,4-triazole was still 31, 12 and 2 % of AR, respectively, in the three soils.

In the model calculations performed for determination of $PEC_{groundwater}$ (according to the data in the EFSA LoEPs for the mentioned azole fungicides and the herbicide amitrole), a formation fraction of 90-100% for 1,2,4-triazole has generally been used as one of the basic parameters for the modelling. This seems plausible as the 1,2,4-triazole moiety in all cases, i.e. all the azole fungicides, are only attached to the rest of the molecule at one point via a C-N bond, which presumably rather easily can be cleaved, predominantly resulting in a free 1,2,4-triazole molecule.

If, as worst case, it is assumed that:

- (i) 100% of the applied standard dose rate hits the soil surface,

- (ii) full formation of 1,2,4-triazole (100%) takes place when an azole fungicide molecule is degraded in soil,

- (iii) 50% of the metabolite remains after one week (i.e. at the end of the fast part of the biphasic degradation), and

- (iv) this fraction is transported by leaching below the topsoil layer,

then half of the "field rate" potential of 1,2,4-triazole presented in Table 4-3 will be potentially available for groundwater contamination, i.e.:

- 11-13 gram/ha (1,1-1,3 mg/m²) for difenoconazole, epoxiconazole and propiconazole
- 28-30 gram/ha (2,8-3,0 mg/m²) for tebuconazole, triadimefon and triadimenol
- 1950 gram/ha (195 mg/m²) for amitrole (probably with considerable variation)

Today, the agricultural uses of azole fungicides are only permitted during spring and summer (until 31 July) but earlier, autumn applications were also approved. With regard to amitrole the main part of its use, which terminated in the late 1980'ies, is assumed to have taken place in the autumn as the substance was mainly used in stubble fields for post-harvest control of couch grass. Amitrole was also used pre-blooming and post-harvest in fruit plantations. The uses in forestry may have taken place from early spring to late autumn.

The autumn applications are the most critical in relation to risk of leaching and subsequent groundwater contamination with 1,2,4-triazole due to surplus of rain in contrast to the summer period. In a historical perspective, autumn application appears to have been the most widespread practice for the herbicide amitrole but also to have had some significance until rather recently for the fungicides used in cereals and other field crops.

5 Azole fungicides for preservation of wood

The azole fungicides used for preservation of wood in Denmark are propiconazole and tebuconazole, most often in combination with IPBC and/or other biocides (insecticides), and in some cases with each other. Many commercial products from a number of suppliers are or have been on the market including products for both industrial wood preservation and for private application by brush or similar (retail sale). It is assumed that the majority of the consumption is associated with the sale of products for industrial use.

Specific data on the consumption for this category of use is only available for the period 2007-2016 while in earlier years only the sum of sales for pesticidal and biocidal uses has been published. The sales data for the biocidal use of propiconazole and tebuconazole for preservation of wood are shown in Table 5-1.

Other biocidal uses of azole fungicides than preservation of wood are described in chapter 8 (Industrial chemicals containing 1,2,4-triazole).

Table 5-1 Consumption of azole fungicides used for preservation of wood in Denmark, 2007-2016, in kg a.s. and in % of total sales (pesticide + biocide).

Year	Propiconazole		Tebuconazole	
	Kg a.s.	% of total	Kg a.s.	% of total
2007	9234	52	6165	23
2008	6160	24	3195*	8.0
2009	6365	38	2324*	5.8
2010	4525	36	2032	4.3
2011	3324	22	1669	3.4
2012	5070	30	1311	2.2
2013	3836	23	1205	1.5
2014	4902	38	1649	4.5
2015	5842	52	1271	2.8
2016	6186	68	1574	2.6
Total	55,444		22,395	

* Calculated as total minus agricultural field pesticides and therefore includes small amounts used for seed treatment and for non-professional use (i.e. pesticide uses).

Additionally, it can be deducted from Table 4-4 that the total consumption of propiconazole and tebuconazole for wood preservation purposes in Denmark from 2000-2017 was 167,111 kg a.s. and 252,549 kg a.s., respectively. The majority of the total consumption for this biocidal purpose was before 2007.

Based on review of the product sheets for a few industrial and domestic products containing azole fungicides, it appears that the content of the fungicides is at about the same level irrespective of the type of product (industrial or domestic). Often two or three substances are used in combination (typically, propiconazole, tebuconazole and IPBC (3-iodo-propynyl-butyl carbamate). The content of propiconazole in the products is typically 0.5-0.9% and 0.2-0.5% in tebuconazole products.

For products (domestic or professional) applied on a wooden board with the same dimensions using a brush, the standard spreading rate is 8 m^2/liter product. This corresponds to about 0.0063 kg azole fungicide/m^2 of treated wood (assuming 0.5% azole and that 1 liter = 1 kg).

For industrial pressure impregnation, some 25-30 litres (or kg) product per m^3 of wood are typically prescribed. With an azole concentration of 0.5% this corresponds to about 0.125-0.150 kg azole fungicide/m^3 wood. It is important to note that impregnation results in penetration of the fungicide deeper into the wood than a surface treatment and, hence, the azole concentration available for wash-off at the surface is probably about the same as for surface-treated wood.

Preserved wood, whether surface-treated or impregnated, is most often used in environments exposed to permanent humidity or to rain or frequent applications of water. I.e. outdoor uses such as surfaces of wooden houses, sheds and carports, wooden fences and poles, wooden bridges and/or harbour constructions. In some cases, the impregnated wood will afterwards be coated with paint to obtain a certain colour or surface appearance, which will reduce the direct exposure of the impregnated wood to rain etc.

The mentioned uses are presumably typically located in urban or residential areas, or near farmhouses, which may also to some extent be placed in groundwater abstraction areas. The gradual release of azole fungicides, e.g. due to wash-off by rain, thereby becomes potentially relevant in terms of risk of groundwater contamination.

However, the release of 1,2,4-triazole from these uses will take place slowly and even when the treated wood is disposed of as waste after several years of use, it will still contain a significant fraction of the originally applied amount of azole fungicide. In Denmark, such wooden waste must be handed in at recycling stations to be incinerated in large-scale municipal solid waste incinerators, although minor amounts might still be burnt in domestic stoves. Hereby, the remaining content of azole substances will be destructed.

6 1,2,4-triazole as nitrification inhibitor

The REACH registration dossier for 1,2,4-triazole indicates that 1,2,4-triazole is produced in or imported into the EU in quantities of 1,000 – 10,000 t/year by three active registrants. The only application specifically mentioned, apart from manufacture of other chemicals, is the use as nitrification inhibitor and as fertiliser.

Liquid nitrification inhibitor

When used as nitrification inhibitor, 1,2,4-triazole is applied only in combination with other nitrification inhibitors such as DCD (dicyandiamide) or 3-MP (3-methyl-pyrazol). The main advantage is a significant synergistic effect with other inhibitors.[2]. Nitrification inhibitors makes it possible to spread organic fertilisers early in the spring while it is still cold and wet without the risk of N-losses from the nitrate form. In Denmark it is particularly used on sandy soils.

The German manufacturer of fertilisers, and one of the three REACH registrants for 1,2,4-triazole, SKW Stickstoffwerke Piesteritz GmbH, manufactures the following products with 1,2,4-triazole:

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2 https://www.fertilizer.org/images/Library_Downloads/2010_Trenkel_slow%20release%20book.pdf

- Piadin®[3]: 1,2,4-triazole + 3MP
- ALZON® Liquid-S 25/6[4]: 1,2,4-triazole + DCD

According to the manufacturer, PIADIN® delays the conversion from ammonium to nitrate. Conversion is, however, not inhibited completely at any point. As a result, the plant can feed itself from both forms of nitrogen in line with requirements at all times.

Piadin® is a liquid solution and contains according to the safety data sheet 2.7-2.9% 1,2,4-triazole[5] in combination with 3-methylpyrazol (3-MP). In Denmark, Piadin was marketed for a few years until 2014. The product was marketed by Biocover in cooperation with Brenntag [6]. According to the importer, the total quantity imported was a few m^3 per year. With a 1,2,4-triazole concentration of about 2.8%, the total 1,2,4-triazole content has been 50-100 kg and compared to the use in pesticides, the consumption has thus been very limited. Besides this import, some direct import took place by a major supplier of products for the agriculture. Piadin® is not part of the standard product range of the company, but Piadin was imported on request from some customers. It has not been possible to obtain information on quantities imported by the company.

A number of different nitrification inhibitors are used in Denmark. According to the available information, the main products are Vizura® based on 3,4-dimethyl-1H-pyrazol and phosphoric acid and N-lock based on nitrapyrin. None of these contain 1,2,4-triazole.

Solid fertilisers

Some fertilisers may also contain 1,2,4-triazole. As an example, Alzon® 46, is a stabilised nitrogen fertiliser for the UK market manufactured by SKW Piesteritz [7]. The product is an ammonium fertiliser, containing a 1,2,4-triazole based nitrogen stabiliser, which is incorporated during manufacture. The nitrogen stabiliser within Alzon® 46 controls the fertilisers change from ammonium to nitrate, allowing higher application rates without increased risk of nitrate losses. This provides crops with a source of ammonium which converts into nitrate in a controlled manner throughout the growing season.

The Danish Agricultural Agency is regularly preparing a list of producers and distributors who import or produce fertilisers, soil improvers, and similar products. The inventory provides an overview of notified products. According to the Danish Agricultural Agency, it is obligatory to indicate if the fertilisers contain nitrification inhibitors. Two fertilisers which contain nitrification inhibitors

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[3] http://www.piadin.de/#home

[4] https://www.skwp.de/fileadmin/user_upload/02_produkte/022_agrochemie/0221_dokumente_deutsch/alzonfluessigs25_6_deu.pdf

[5] http://cdn.i-pulse.nl/triferto/userfiles/1/pdf%20NED/piadin-msds-triferto-nl.pdf

[6] http://www.biocover.dk/images/pdf/PIADIN_dansk.pdf

[7] https://www.gleadell.co.uk/uploads/gleadell-Alzon-46-brochure.pdf

are currently listed in the fertiliser inventory (Danish: Gødningsfortegnelse fra Landbrugsstyrelsen) [8]. None of these contain 1,2,4-triazole. Alzon® 46 is not listed. On this basis, it is estimated that fertilisers with 1,2,4-triazole based inhibitors are not produced or legally imported into Denmark.

7 Medicinal products with 1,2,4-triazole

7.1 Medicinal products against fungal infection

According to the medicinal products database, 5 antifungal agents containing a 1,2,4-triazole ring may be used in in Denmark (Lægemiddelstyrelsen, 2018). The substances are listed in the table below which also indicate the percentage of the substances accounted for by the 1,2,4-triazole moieties. The percentage is used to calculate the potential formation of 1,2,4-triazole from degradation of the substances.

Table 7-1 Identified antifungal agents which may be degraded to 1,2,4-triazole.

Pharmaceutical name	Chemical name	CAS No	Molecular weight	1,2,4-triazole moiety as % of total
Isavuconazole	4-[2-[(1R,2R)-2-(2,5-Difluorophenyl)-2-hydroxy-1-methyl-3-(1H-**1,2,4-triazol**-1-yl)propyl]-4-thiazolyl]benzonitrile	241479-67-4	437.47	16%
Fluconazole	a-(2,4-Difluorophenyl)-a-(1H-1,2,4-triazol-1-ylmethyl)-1H-**1,2,4-triazole**-1-ethanol	86386-73-4	306.27	45% (two moieties)
Itraconazole	(+/-)-4-[4-[4-[4-[[(2R,4S)-(2,4-Dichlorophenyl)-2-(1H-1,2,4-triazol-1-ylmethyl)-1,3-dioxolan-4-yl]methoxy]phenyl]-1-piperazinyl]phenyl]-2,4-dihydro-2-(1-methylpropyl)-3H-**1,2,4-triazol**-3-one; Itrizole	84625-61-6	705.63	10%
Voriconazole	2R,3S-2-(2,4-Difluorophenyl)-3-(5-fluoropyrimidin-4-yl)-1-(1H-**1,2,4-triazo**l-1-yl)butan-2-ol	137234-62-9	349.31	20%
Posaconazole	2,5-Anhydro-1,3,4-trideoxy-2-C-(2,4-difluorophenyl)-4-[[4-[4-[4-[1-[(1S,2S)-1-ethyl -2-hydroxypropyl]-1,5-dihydro-5-oxo-4H-1,2,4-triazol-4-yl]phenyl]-1-piperazinyl]phenoxy]methyl]-1-(1H-**1,2,4-triazol**-1-yl)-D-threo-pentitol	171228-49-2	700.78	10%

A review of triazole antifungals (Peyton *et al.*, 2015) further lists the following 1,2,4-triazole based antifungals, which according to the statistics was not sold in Denmark in 2017: Albaconazole, Efinaconazole, and Ravuconazole. Besides the antifungal agents based on 1,2,4-triazole, a number of agents based on other azoles are marketed.

[8] https://lbst.dk/fileadmin/user_upload/NaturErhverv/Filer/Landbrug/Handelsgoedning/Goedningsfortegnelse_03.07.17.pdf

Data on the consumption of the antifungal agents have been extracted from the Danish Register of Medicinal Products Statistics (in Danish: Lægemiddelstatistik-registret) of the Danish Medicines Agency. The register contains information on all medicinal products sold in Denmark. The public database of the register is available at http://medstat.dk/.

Data for the five substances, as extracted from the register, are shown in the table below. The data are for antifungal agents in the register indicated as defined daily dose (DDD). The DDD is a standard daily dose established by the WHO for each medicinal product and route of administration. The majority of the agent are for oral administration and the DDD for oral administration is used for the calculations.

Table 7-2 Number of registered daily doses for the antifungal agents and total content of 1,2,4-triazole moieties.

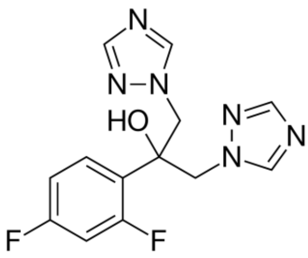
Pharmaceutical name	**Number of DDD (defined daily doses) in 2017, approx. *1**	**DDD oral administration, mg *2**	**Total supply in 2017, kg 1,2,4-triazole *3**
Isavuconazole	1,700	200	0.1
Fluconazole	925,000	200	83
Itraconazole	210,000	200	4
Voriconazole	26,000	400	2
Posaconazole	53,000	400	2
Total			91.8

*1 Register of Medicinal Products Statistics at: http://medstat.dk/

*2 Source: WHO ATC/DDD Index 2018 at: https://www.whocc.no/atc_ddd_index/. DDD for oral administration.

*3: Total supply = number of DDD per year * DDD * percentage 1,2,4-triazol moiety (the latter from Table 7-1)

The main consumption is with fluconazole which have two 1,2,4-triazole moieties.



Structural formula of fluconazole

According to the medicinal products database pro.medicin, about 80% of the fluconazole is excreted through the kidneys as unchanged substance and no metabolites are identified [9].

The pharmacokinetics for itraconazole is very different, as the renal excretion as unchanged substance is only 0.03%, whereas 3-18% is excreted with faeces and 35% is excreted as metabolites through the kidney[10] (not indicated to what extent the metabolites contain a 1,2,4-triazole moiety).

Only 2% of the voriconazole is excreted through the kidney and the substance is reported to be metabolised in the liver to inactive metabolites[11].

The majority of posaconazole is excreted with faeces as unchanged substances [12].

In summary, fluconazole represents the major part of the consumption and with a renal excretion of 80% of the substance, it represents close to 100% of the 1,2,4-triazole discharged to the waste water system from the five mentioned substances. In total, in 2017 the quantities discharged would be approximately 80% of 85 kg (sum of fluconazole and posaconazole) used corresponding to approximately 68 kg of 1,2,4-triazole.

Kahle *et al.* (2008) present data on fluconazole in influent to sewage treatment plants in Switzerland. The mean load is estimated at 0.028 mg/person/day. If the same would be the situation in Denmark, the total discharges to sewage treatment plants would be 57 kg/year which is lower than the estimated 157 kg/year in 2017 (corresponding to 66 kg 1,2,4-triazole) but still in the same order of magnitude. The majority of the fluconazole in the Swizz sewage treatment plants was discharged with the effluent from the sewage treatment plants and the average discharges corresponded to 0.016 mg/person/day (57% of influent) (Kahle et al, 2008). The study found that the fluconazole was not degraded by activated sludge incubation. The study does not report the concentration of fluconazole in the sludge.

In accordance with the results from Switzerland, Peng *et. al* (2012) found that fluconazole passed through treatment in sewage treatment plants in China and largely remained in the final effluent. The study did not analyse for fluconazole in the sludge because previous studies had not found fluconazole in the sludge.

In a study from Sweden, Lindberg *et al. (*2010) found that negligible amounts of fluconazole were removed from the aqueous phase, and its levels were below the limit of quantification in all of the analysed sludge samples. The study concluded that 53% of the total amount of fluconazole sold in Sweden appeared in the final effluents of the sewage treatment plants (Lindberg et al., 2010). The

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[9] Fluconazol "HEXAL". https://pro.medicin.dk/Medicin/Praeparater/3228#a110
[10] Sporanox® Itraconazol.
https://pro.medicin.dk/Medicin/Praeparater/1444#a110
[11] Voriconazole "Sandoz".
https://pro.medicin.dk/Medicin/Praeparater/8151#a110
[12] Noxafil® Posaconazol.
https://pro.medicin.dk/Medicin/Praeparater/3813#a400

other antifungal agents studied, terbinafine, clotrimazole, ketoconazole, econazole and miconazole, do not include a 1,2,4-triazole moiety.

If it as a worst case is assumed that the remaining 43% ends up in the sludge (but more likely it is significantly lower), the 68 kg 1,2,4-triazole discharged to sewage treatment plants in Denmark would correspond to 29 kg ending up in the sludge. In Denmark about 60% of the sewage sludge is used for agricultural purposes and consequently approximately 18 kg 1,2,4-triazole may have been applied on agricultural soils from the use of fluconazole. By degradation of the fluconazole, the 1,2,4-triazol group may be released.

7.2 Other medicinal products

A search in the medstat.dk database for substances listed in Kaur et al. (2017), Wikipedia "1,2,4-triazole derived medicinal products" (14 products), and non-cancer agents mentioned at pro.medicin.dk have revealed six other medicinal products which contain a 1,2,4-triazole moiety. The substances are used for various applications as shown in the table below.

Table 7-3 Identified other medicinal products which may be degraded to 1,2,4-triazole.

Medicinal product name	**Chemical name**	**CAS No**	**Molecular weight**	**1,2,4-triazole moiety as % of total**	**Application**
Triazolam	8-chloro-6-(2-chlorophenyl)-1-methyl-4H-**[1,2,4]triazolo**[4,3-a][1,4]benzodiazepine	28911-01-5	347.23	20%	Treatment of insomnia
Alprazolam	8-chloro-1-methyl-6-phenyl-4h-**[1,2,4]triazolo**[4,3-a][1,4]benzodiazepine	28981-97-7	308.76	22%	Sedative
Rizatriptan	n,n-dimethyl-2-[5-(**1,2,4-triazol**-1-ylmethyl)-1h-indol-3-yl]ethylamine benzoate salt, antimigraine	145202-66-0	391.47	18%	Antimigraine
Letrozole	4,4'-(1h-**1,2,4-triazol**-1-ylmethylene)bisbenzonitrile	112809-51-5	285.30	24%	Aromatase inhibitor for breast cancer
Anastrozole	2;2"-[5-(1h-**1,2,4-triazol-**1-ylmethyl)-1,3-phenylene]bis(2-methyl-propiononitrile	120511-73-1	293.37	24%	For estrogen receptor-positive breast cancer
Ribavirin	1-β-D-ribofuranosyl-**1,2,4-triazole**-3-carboxamide	36791-04-5	244.20	28%	Antiviral agent for chronical hepatitis C

The total use of the substances in 2017 is shown in the table below. The total supply in 2017 of 1,2,4-triazole equivalents was about 2 kg and the total for these other substances is small as compared to the supply with the antifungal agents. The pharmacokinetics of the substances has not been assessed but the total 1,2,4-triazole equivalent excreted would be lower than 2 kg.

Table 7-4 Number of registered daily doses for the other medicinal products and total content of 1,2,4-triazole moieties

Pharmaceutical name	**Number of DDD (defined daily doses) in 2017, approx. *1**	**DDD, mg *2**	**Total supply in 2017, kg 1,2,4-triazole *3**
Triazolam	211,000	0.25	0.04
Alprazolam	3,142,000	1	0.67
Rizatriptan	640,000	10	0.78
Letrozole	109,000	2.5	0.07
Anastrozole	314,000	1	0.07
Ribavirin	18,500	6	0.03
Total other			2.01

*1 Register of Medicinal Products Statistics at: http://esundhed.dk/sundhedsregistre/LSR/Sider/MEDQTR-Diagram.aspx

*2 Source: WHO ATC/DDD Index 2018 at: https://www.whocc.no/atc_ddd_index/. DDD for oral administration.

*3: Total supply = number of DDD * DDD * percentage 1,2,4-triazol moiety

According to a review of the recent developments on 1,2,4-triazole nucleus in anticancer compounds, 1,2,4-triazole is an important nucleus present in a large number of compounds (Kaur et al., 2016). More than thirty-five compounds containing this nucleus are introduced into the market. According to Kaur et al. (2016), 1,2,4-triazole nucleus is stable to metabolism and acts as an important pharmacophore by interacting at the active site of a receptor as hydrogen bond acceptor and as a donor. Due to its polar nature, the triazole nucleus can increase the solubility of the ligand and it can significantly improve the pharmacological profile of the drug. Data for about 20 non-cancer compounds identified at pro-medicin.dk has been retrieved from the medicinal products statistics but none of the substances with reported use contained a 1,2,4-triazol moiety.

The list of substances identified in the medicinal products register may not be comprehensive and not contain all substances with 1,2,4-triazole moiety listed in the Danish Register of Medicinal Products Statistics. The data, however, clearly indicate that the contribution from medicinal products to the total releases of 1,2,4-triazol to agricultural soils is very small as compared with other sources.

Two other substances have been identified in the literature and via the internet: Etizolam and Furacylin; no use was registered in the medicinal products statistics.

On 16 December 2014, orphan designation (EU/3/14/1392) was granted by the European Commission to Palobiofarma S.L., Spain, for 5-bromo-N-(prop-2-yn-1-yl)-2-(1H-1,2,4-triazol-1-yl)pyrimidine-4,6-diamine for the treatment of Huntington's disease. An orphan drug is a pharmaceutical agent that has been developed specifically to treat a rare medical condition, the condition itself being referred to as an orphan disease. The total consumption has not been further assessed as it is assumed to be insignificant.

A wealth of information is available in the scientific literature regarding the possible use of medicinal products with 1,2,4-triazole. A search in the bibliographic database PubMed gave 1072 hits for the text string "1,2,4-triazole" in the title of the publications. The literature has not been reviewed as part of the current study.

8 Industrial chemicals containing 1,2,4-triazole

As described above, the REACH registration dossier for 1,2,4-triazole indicates that 1,2,4-triazole is produced in or imported into the EU in quantities of 1,000 – 10,000 t/year by three active registrants. The descriptions of applications are in general very generic (e.g. formulation of fine chemicals). The only application specifically mentioned are the uses as nitrification inhibitor and as fertiliser.

The German chemical manufacturer, Lanxess (business unit Saltigo), one of the three registrants of 1,2,4-triazole, indicates the following applications for 1,2,4-triozole: Agriculture, building block for agrochemicals, building block for pharmaceuticals and fine chemicals, and pharmaceutical industry/biotechnology. These applications are all covered by other chapters in this note.

A large number of substances contain the 1,2,4-triazole moiety. A search in Classification and Labelling Inventory (C&L inventory) at ECHA's website of substances with the text string "1,2,4-triazole" resulted in a list about 800 substances with a self-classification. About 1,000 substances with the text string "1,2,4-triazole" in the chemical name are pre-registered under REACH and 92 substances are registered.

The SPIN database of the Nordic product registers contains in total 56 substances with the text string "1,2,4-triazol" in the chemical name. Data for these 56 substances have been retrieved from the Danish Product Register, December 2018. The data are not shown for reasons of confidentiality.

For substances where non-confidential data are available in the Danish Product Register, data have been extracted from the SPIN database[13] for the most recent year 2016 and shown in the table below. The table does not include agricultural pesticides.

The data in the Danish Product Register represent chemical products (mixtures) for professional use imported or produced in quantities above 100 kg/year. The

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[13] SPIN - Substances in Preparations In the Nordic countries - is a database that contains "non-confidential" information on substances from the product registers of Norway, Sweden, Finland and Denmark. http://spin2000.net/

duty to declare the products applies to products with at least one constituent with a harmonised classification, but in this case all constituents are declared. Products with a biocidal function always have to be declared. Foodstuffs, medicinal products and cosmetic products are exempted for the duty to declare products. It means that some substances with a 1,2,4-triazole moiety, which do not have a harmonised classification or are used exclusively in products for consumer applications, may not be declared to the Danish Product Register.

The main uses in 2016 are 4.6 t/year tebuconazole and 5.2 t/year propiconazole used for the UC62[14] categories "non-agricultural pesticides and preservatives" and "impregnation materials". When the data are represented using Danish national use codes, the only listed applications are "Wood impregnation agents, wood preserving agents (see also Pesticides)" and "Wood preservatives" but the listed quantities are lower than the quantities shown when the data are represented using UC62 codes. These applications are included in chapter 5 on fungicides used for preservation of wood.

As it appears from Table 8-1 below, a total of 0.4 t/year are used in paints, lacquers and varnishes, and 0.1 t/year lubricants and additives.

The function of tebuconazole and propiconazole in paints, lacquers and varnishes may be as a film preservative (PT 7) as both substances are approved for this application, but they are not approved for use as in-can preservative. The substances are also registered for use in paints, lacquers and varnishes in other Nordic countries. A search for Safety Data Sheets (SDSs) for products with tebuconazole or propiconazole on the Danish market mainly identified SDSs for wood preservatives. A few SDSs for outdoor wood oil, outdoor paint based on linseed oil and a solvent-based outdoor paint were identified[15]. The concentration of the substances are typically 0.1-1 % i.e. the same concentration as in wood preservatives. The substances may be released to the environment from this application by the same mechanisms as described for wood preservatives, but the quantities used are significantly lower than the quantities used for wood preservatives.

In addition, one example of a product witt propiconazole for general construction material preservation including wood [16] (propiconazole is not approved for PT10 - Construction material preservatives) and one example of a silicone sealant with Tebuconazole [17] were identified. The quantities used are not reported in SPIN and are likely small.

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[14] UC62 is a common use categories system used across the Nordic product registers with 62 categories.

[15] Two examples of SDSs of outdoor paints made of linseed oil: https://www2.nst.dk/apb/LBA/Damson/Damson_Laserende_Linoliemaling_DA_2.0.pdf; https://m.carl-ras.dk/public/dokumenter/catalog/25738_D_49927609_Produktinformation_1333321__25743_D_49927609_Sikkerhedsdatablad_1333321.pdf

[16] http://www.byggecenter.dk/uploads/product_files/20110216171132_protoxskimmel_tmb.pdf

[17] http://www.pattex.dk/content/dam/uac/pattex-responsive/denmark/movedAssets/pattex/denmark/msds/pattex-mouldblocker--all-colours-sds413833-140527-dk.pdf

The application of 1-((bis(2-ethylhexyl)amino)methyl)-1,2,4-triazole in lubricants and hydraulic fluids are also reported in SPIN for the other Nordic countries, but in similar small quantities. An example of the additive product is the Metal Deactivator - IRGAMET® 30 from BASF, which is used in concentrations of 200-1000 ppm in industrial lubricants and 10-50 ppm in transformers. [18] The function is to protects metal from corrosion. The releases from these uses are likely small, as the waste oils and lubricants are mainly disposed of as hazardous waste. Considering the small total quantities used, the possible release pathways have, however, not been further assessed.

The Danish Product Register does not include consumer products, but the Swedish Product Register does also include such products. For the substances listed in the table below, the data in SPIN from the Swedish Product Register are not markedly different from the Danish data, and the available data do not point at any significant use in consumer products not covered by the Danish Product Register.

Table 8-1 Uses of substances with 1,2,4-triazole in the chemical name as registered in the SPIN database for 2016

Name/trivial name	**CAS No**	**UC62 use category**	**No of products**	**Tonnage t/year**
Tebuconazole	107534-96-3	Non-agricultural pesticides and preservatives	7	3.2
		Impregnation materials	4	1.4
		Paints, lacquers and varnishes	7	0.3
1,2,4-triazole	288-88-0	No uses indicated	-	-
Propiconazole	60207-90-1	Non-agricultural pesticides and preservatives	17	4.5
		Impregnation materials	14	0.7
		Paints, lacquers and varnishes	27	0.1
1h-1,2,4-triazole, 1-((2-(2,4-dichlorophenyl)-4-ethyl-5-methyl-1,3-dioxolan-2-yl)methyl)-	75296-54-7	No uses indicated	-	-
1-((bis(2-ethylhexyl)amino)methyl)-1,2,4-triazole	91273-04-0	Lubricants and additives	9	0.1
		Hydraulic fluids and additives	13	0.0

18 http://www.performancechemicals.basf.com/ev/internet/lubricants/en/content/lubricants/products/metal_deactivators/irgamet_30

9 Other possible sources of 1,2,4-triazole

Natural formation in the environment

Substances with a 1,2,4-triazole moiety may be produced by natural processes in the environment. Examples are essramycin which was isolated from a marine Streptomyces species from the Egyptian Mediterranean coast (Wang et al., 2016) and 1,2,4-triazole-3-alanine isolated from actinomycetes from a Japanese soil (Imamura et al., 1985).

Very limited information is available regarding the contribution of naturally formed substances with 1,2,4-triazole to the 1,2,4-triazole levels found in the environment.

The monitoring results of 1,2,4-triazole from the Danish Pesticide Leaching Assessment Programme from Estrup, Faardrup, Tylstrup and Jyndevad reveal (except for Faardrup) an unprecedented high background concentration level of 1,2,4-triazole in water samples collected from 1 m depth and in groundwater. (Rosenbom, 2016). The level declined with depth, which according to the authors indicates a source coming from the field surface. With the background concentration level it is, however, according to the authors clear that the source resulting in the many detections can not only be the tebuconazole application, but an outcome of earlier applications of fungicides having 1,2,4-triazole as a degradation product or even other sources (Rosenbom, 2016).

Three conference proceedings in a series "The triazole story" from 2018, authored in cooperation between some major manufacturers of agricultural chemicals and the German SGS Institute Fresenius (testing and inspection services), addresses the importance of other sources than agricultural chemicals. It should be noted that conference proceedings are usually not peer reviewed.

Blank et al. (2018a) have studied the differentiation between different 1,2,4-triazole sources using a ^{13}C stable isotope labelled azole-fungicide. In the study, ^{13}C-labelled tebuconazole (^{13}C-TBZ) was applied once to bare soils in six different locations across Europe (Spain, Italy, UK, Germany, Belgium, and Denmark). The use of non-labelled triazole fungicides or N-stabilized fertilisers could be excluded for all sites since 2013 i.e. for three years. Soil samples were collected at 15 sampling times from 0 to 360 days after application, in triplicates and in additional control plots. The soil specimens were analysed for residues of ^{13}C labelled and natural ^{12}C tebuconazole and 1,2,4-triazole (124 T).

The paper present data from the soils in Denmark and Spain. In the Spanish soils the level of ^{13}C 1,2,4-triazole peaks at a level of 4 g/ha while the level of ^{12}C 1,2,4-triazole is at 0 g/ha. In the Danish soils the level of ^{12}C 1,2,4-triazole is far above the level of ^{13}C 1,2,4-triazole as shown in the figure below with a peak level of 120 g/ha for one of the soils. Data from the other countries are not presented and it is not clear if the results are similar to the results from Spain.

The authors conclude that the results confirm that the origin of the 1,2,4-triazole in the Danish soils is not the application of the fungicides. However, the data presented do not exclude that the source of the 1,2,4-triazole in the Danish soil is the former use of 1,2,4-triazole fungicides used historically and accumulated in the soils. Further, it is strange that data from four of the countries have been excluded from the paper without explanation.

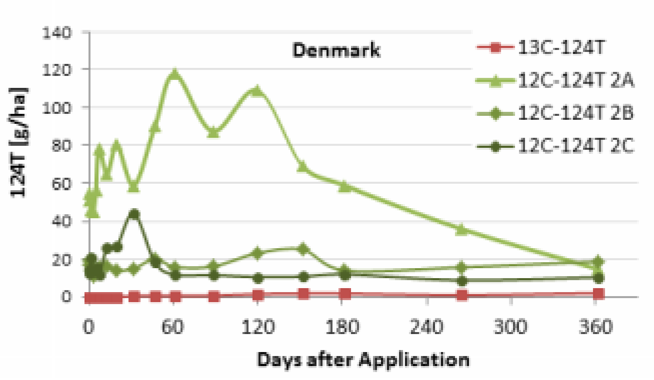

Figure 9-1 *^{13}C and ^{12}C in Danish soils after application of 12C labelled a ^{13}C stable isotope labelled azole-fungicide (from Blank et al., 2018a).*

Blank et al. (2018b) report 1,2,4-triazole levels in forest soils from a field study of forest top soil samples from 10 different locations and different forest types (beech, spruce, pine, oak) in Germany sampled in 2012/201. The background abundance of 1,2,4-triazole in the samples ranged from <1.0 to 1.9 µg/kg in oak forest top soils, from 1.0 to 2.1 µg/kg in pine forest top soils, and from <1.0 to 1.2 µg/kg in spruce forest top soils. In the selected beech forest top soils, the background abundance of 1,2,4-triazole was below 1.0 µg/kg. A simulation of the forest soils was run to describe a potential formation of 1,2,4-triazole in forest soils as a natural process, without any pesticide application. Three different "assumptions/ fits" was used. The results for all resulting 6 calculations of the 1,2,4-triazole formed ranged between 28.3 and 37.3 g/ha*year (mean 32.8 g/ha*year).

Blank et al. (2018c) summarise data on 1,2,4-triazole in German standard soils, which contained 1,2,4-triazole in concentrations up to 2.29 µg/kg dry soil. Forest soils from ten locations in Germany contained up to 2.1 µg/kg dry soil (with reference to unpublished data of Naeb (2017), but apparently the right reference is unpublished data by Heinemann (2013)). Furthermore, the paper notes that soils from a field study in six locations across Europe using ^{13}C labelled tebuconazole showed unlabelled 1,2,4-triazole concentrations up to 120 g/ha (apparently the same data as shown above but with another reference). The authors conclude that "*The work has shown that there are a wide range of sources of 124T...*". However, without comparing background levels with levels in agricultural soils with application of 1,2,4-triozole fungicides, the study does not indicate the significance of other sources in comparison with the fungicides.

Thus, the annual natural formation of 1,2,4-triazole in soil seems to be at about the same level as the annual "1,2,4-triazole potential" input to agricultural soils from the application of azole fungicides in field crops, which range from 21 to 60 g/ha*year for the different active fungicidal azole substances (see section 4.2.2). However, the natural formation takes place more or less throughout the year (probably less during winter time) while the agricultural application is made at one specific point in time, which could indicate a higher risk of leaching of the agricultural 1,2,4-triazole compared to the natural formation where degradation of the substance takes place continuously parallel with formation of new 1,2,4-triazole. Moreover, it is not clear from the data if natural formation also occurs in agricultural soils or if it is confined to forest soils.

A search in Pub.Med with "1,2,4-triazole" AND "environmental" (or "environment") did not reveal other studies of the natural environmental formation of 1,2,4-triazole, but a number of articles in environmental degradation. A search for similar articles or articles citing Wang et al. (2016) and Imamura et al. (1985) did not result in any articles addressing 1,2,4-triazole formation.

Explosives

1,2-dihydro-5-nitro-3H-1,2,4-triazol-3-one (CAS No 932-64-9) is registered under REACH with a production/import in the 100-1000 t/y range. The applications are indicated as production of explosive items, ammunition, and pyrotechnic devices.

A number of scientific papers regarding the use of 3-nitro-1,2,4-triazole-5-one (synonym: 3-nitro-1,2,4-triazole-5-one).

According to Khatiwada et al. (2018) "*The emerging insensitive munitions compound (IMC) 3-nitro-1,2,4-triazole-5-one (NTO) is currently being used to replace conventional explosives such as 1,3,5-trinitro-1,3,5-triazacyclohexane (RDX), but the environmental fate of this increasingly widespread IMC remains poorly understood. Upon release from unexploded solid phase ordinances, NTO exhibits high aqueous solubility and, hence, potential mobilization to groundwater. Insensitive munitions compounds (IMCs) are increasingly used for military energetic materials, yet their environmental fate is poorly understood. Prior work has shown that the nitroaromatic 2,4-dinitroanisole (DNAN) and the heterocyclic nitrogen compound 3-nitro-1,2,4-triazole-5-one (NTO), both newly introduced IMCs, can undergo microbially mediated reduction under anoxic conditions to form 2-methoxy-5-nitroaniline (MENA) and 3-amino-1,2,4,triazole-5-one (ATO) respectively*".

Mark et al. (2016) studied adsorption and attenuation behaviour of the explosive compound 3-nitro-1,2,4-triazol-5-one (NTO) from explosives in eleven soils with the aim of evaluate NTO potential for natural attenuation in soils. The study did not provide any conclusions regarding the fate in soil.

The possible use of this type of explosives in Denmark and the potential for environmental pollution e.g. at military training grounds have not been further investigated. If the use in Denmark would be at about 1% of the REACH registered volume (if the per capita consumption was at the EU average), the consumption would be in the 1-10 t/y range.

Other possible sources

As mentioned elsewhere, the REACH registration database contains 92 registered substances with "1,2,4-triazole" in the chemical name, and it cannot be excluded that some may be used for applications not included above.

10 Summary and conclusions

Agricultural pesticides. In Denmark, this category comprises 12 active azole substances of which 11 are fungicides while the last one is a herbicide, amitrole.

Amitrole, which consists of 82% 1,2,4-triazole, was used in significant amounts from 1958 to 1989 and was applied to soil at much higher rates than any of the azole fungicides i.e. between 1 and 9.5 kg a.s./ha compared to 0.125-0.250 kg a.s./ha for the azole fungicides. However, considering that the sale stopped 30 years ago (1989), it would be expected that by now all 1,2,4-triazole resulting from this use was completely degraded in the environment.

Propiconazole, of which 20% is 1,2,4-triazole, is by far the azole substance with the highest total consumption over the years, almost 2 million kg a.s., of which probably almost 1,8 million kg has been used in agriculture. The sales of tebuconazole and epoxiconazole for agricultural use amount both to more than 600,000 kg a.s. while the total sales of bitertanol, prothioconazole, triadimefon and triadimenol are in the range approx. 174,000-463,000 kg a.s.

The 1,2,4-triazole moiety constitutes 17-24% of the azole fungicides and thereby represents the theoretical potential for groundwater contamination with this substance. A worst case estimation of the potential for groundwater contamination with 1,2,4-triazole from agricultural pesticides results in a potential per application of 11-30 gram/hectare for the fungicides and 1950 gram/ha for the herbicide amitrole (with considerable variation due to a large dosage range). Autumn application of the azoles, which is no longer permitted in Denmark (since 2014), is considered to be the most critical in terms of risk of groundwater contamination.

Wood preservation fungicides. At the moment, the only two azole fungicides that are used in Denmark for preservation of wood are propiconazole and tebuconazole. The total amounts sold within this category of use from 2000-2017 are around 172,000 kg a.s. and 167,000 kg a.s. It is assessed that the majority of the total sales for wood preservation in Denmark have taken place from 2000 and onwards. The uses within this category range from industrial pressure impregnation of wood to domestic application of surfaces using a brush.

The typical azole fungicide concentration is about about 0.5% in most of the products. For pressure impregnation of wood it is estimated that about 0.125-0.150 kg/m³ of azole fungicide (of which 20-22% is the 1,2,4-triazole moiety) is used. When azole-containing wood preservation products are applied onto the wood surface using a brush or similar the resulting concentration is estimated to about 0.006-0.007 kg azole fungicide/m² treated wood.

Nitrification inhibitor. 1,2,4-triazole may be present in liquid nitrification inhibitors added to organic fertilisers and in some solid fertilisers. However, the amounts appear to be so limited that this category of use is assessed to be of negligible significance at the national scale but may contribute to the total load of 1,2,4-triazole at specific locations. It has not been possible to confirm if this use of 1,2,4-triazole still takes place in Denmark.

Medicinal products. In total, 11 medicinal products with a 1,2,4-triazole moiety used in Denmark have been identified and consumption data have been extracted from the Danish register of medicinal products. The list may not be comprehensive but is considered to represent the major applications. The antifungal agent fluconazole represents the largest contribution to 1,2,4-triazole in waste water from the use of medicinal products. The product represents about 90% of the total 1,2,4-triazole moieties in used medicinal products and in addition, the substance is only to a limited degree metabolised in the body and the renal excretion is 80% of the intake.

Studies from Switzerland and China demonstrates that the majority of the fluconazole in the influent to sewage treatment plants, pass though the plants and is discharged with the effluent. It is estimated that as a worst case 17 kg/year of 1,2,4-triazole from fluconazole (but likely much lower quantity) may be applied on agricultural soils with sewage sludge while the contribution from other medicinal products is considered insignificant.

Industrial chemicals. According to the REACH registration dossier, the industrial use of 1,2,4-triazole is as intermediate in the production of agrochemicals and medicinal products and the use as nitrification inhibitor described separately above. The SPIN database of the Nordic product registers contains in total 56 substances with the text string "1,2,4-triazol" in the chemical name. Data for these 56 substances have been retrieved from the Danish product register and non-confidential data has been retrieved from the SPIN database. The major non-agricultural applications are the use in wood preservatives described separately above. A total of 0.4 t/year was used as in other types of paint.

Available Safety Data Sheet indicate that the use is likely as film preservative in outdoor paints and wood-oils. The release pathways for these applications would be the same as for the wood preservatives, but the total quantity used is significantly lower. About 0.1 t/year was used in lubricants and hydraulic fluids where it serves to protect metal from corrosion. The releases from this application are thus considered insignificant.

Other possible sources. 1,2,4-triazole may be formed by some natural processes in the environment and it has been demonstrated that 1,2,4-triazole may be formed by some natural strains of microorganisms. The calculated annual formation rate in forest soils seems to be close to the same level as the "1,2,4-triazole potential" per application of the azole fungicides applied to agricultural crops, which indicates that natural formation of 1,2,4-triazole is possibly not just negligible although it is not known if this process only takes place in forest soils or also in agricultural soils.

3-nitro-1,2,4-triazole-5-one is used for explosives with a REACH registered volume in the 100-1000 t/year range for the entire EU. The specific use in Denmark and the risk of contamination of military training grounds has not been investigated.

Overall assessment. Based on the information available or obtained for the current small study, it is assessed that at an overall level the agricultural use of azole pesticides has the highest potential for risk of leaching of 1,2,4-triazole to groundwater in Denmark. In particular, applications in the autumn (and early spring) imply a risk due to low temperatures and precipitation surplus in that period. This practice is now discontinued in Denmark (since 2014) but has been common for years.

At the time of its use, amitrole could be a substance, which has contributed significantly to the environmental load of 1,2,4-triazole due to its very high dose rates in combination with its predominant use as a post-harvest herbicide, i.e. with application in the autumn. However, one would expect that by now, 30 years after the sale of amitrole was discontinued in Denmark, all 1,2,4-triazole resulting from the use of this herbicide would be completely degraded in the environment.

Locally, use of azole fungicides for wood preservation (both during production, outdoor storage and use) and use of 1,2,4-triazole as nitrification inhibitor could have some influence while other sources appear to be of only minor significance or negligible.

Finally, it cannot be excluded that natural formation of 1,2,4-triazole also contributes somewhat to the risk of groundwater contamination.

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