



National Institute for Public Health
and the Environment
Ministry of Health, Welfare and Sport

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from grass and soil to meat of wild cattle
grazing in floodplains in the Netherlands**

RIVM letter report 2021-0142
S. Notenboom et al.



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Colophon

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Synopsis

Transfer of dioxins and dioxin-like PCBs from grass and soil to the meat of wild cattle grazing in floodplains in the Netherlands

In a number of areas between rivers and dykes (floodplains), grazing by wild cattle is used as a form of nature management. Some of these animals are slaughtered to manage the size and composition of the herds. Their meat is then sold as 'wilderness meat'. In 2020, excessive dioxin levels were discovered in the meat of some of the wild cattle as a result of dioxins and dioxin-like PCBs in the grass and soil of floodplains.

In response to this situation, the Dutch National Institute for Public Health and the Environment (*Rijksinstituut voor Volksgezondheid en Milieu* (RIVM)) and Wageningen Food Safety Research developed models to calculate the extent to which these substances end up in the meat of these cattle via grass and soil. These models were also used to estimate how long it takes for levels to fall to the permitted level when the cattle are moved to an area outside the floodplains. The concentrations are lower there.

The models were developed for three types of wild cattle in herds: cattle that do not give milk, cattle that give milk and calves, and calculations were based on measurements on grass and soil in the floodplains in the Beuningen area. Some concentrations in grass were high due to recent flooding in the area; others were lower, which is more common. The concentrations present in grass after flooding are often higher because the sediment left behind in the grass contains dioxins and dioxin-like PCBs. The concentrations measured in the soil were approximately the same in both situations.

Regardless of whether the samples contained higher or lower concentrations, the levels measured in meat of all three types of cattle were higher than those permitted by law. If cattle were feeding calves, the levels fell below those permitted by law, because dioxins and dioxin-like substances are excreted via the milk. After cattle had been moved to a cleaner area, the levels calculated fell too. Levels in adult cattle then fell to below the level permitted within one-and-a-half to four months. This fall in levels took more than six months to achieve among calves.

Dioxins and dioxin-like PCBs are chemical substances that are created during the incineration of waste and other substances. Despite the strong decrease in emissions in the last 25 years, dioxins and dioxin-like PCBs are still present in the Netherlands (in grass, the soil and river sediment, for example). Dioxins can be harmful to the immune system, brain development and reproduction.

Another RIVM and WFSR report describes how the models were made.

Keywords: dioxins, PCBs, PBK-modelling, floodplains, cows

Publiekssamenvatting

Overdracht van dioxines en dioxine-achtige PCB's van gras en bodem naar vlees van wilde runderen die grazen in de uiterwaarden in Nederland

Wilde runderen grazen in enkele gebieden tussen de rivier en dijk (uiterwaarden) als een vorm van natuurbeheer. Sommige dieren worden geslacht om de grootte en samenstelling van de kuddes goed te houden. Het vlees wordt verkocht als 'wildernisvlees'. In 2020 zijn in het vlees van enkele wilde runderen te hoge hoeveelheden dioxinen gemeten. Dit komt door de dioxinen en dioxine-achtige PCB's in het gras en in de grond in uiterwaarden.

Het RIVM en Wageningen Food Safety Research hebben daarom modellen ontwikkeld om te berekenen hoeveel van deze stoffen via gras en grond in het vlees van deze runderen terecht komen. Hiermee is ook geschat hoe lang het duurt voordat de hoeveelheden op het toegestane niveau komen als de runderen naar een gebied buiten de uiterwaarden worden verplaatst. Daar zijn de concentraties lager.

De modellen zijn ontwikkeld voor drie typen wilde runderen in een kudde: runderen die geen melk geven, runderen die melk geven, en kalveren. Voor de berekeningen zijn metingen in gras en bodem in de uiterwaarden in de buurt van Beuningen gebruikt. Er is gerekend met hoge concentraties in gras die vlak na een overstroming zijn gemeten. Daarnaast is met lagere concentraties gerekend, die vaker voorkomen. De concentraties in gras zijn na een overstroming vaak hoger doordat slib dat achterblijft op het gras, dioxinen en dioxine-achtige PCB's bevat. De gemeten concentraties in de bodem waren in beide situaties ongeveer hetzelfde.

Zowel bij de hogere als de lagere concentraties kwamen de berekende hoeveelheden in het vlees van alle drie typen wilde runderen boven de hoeveelheid uit die wettelijk is toegestaan. Wanneer koeien hun kalveren melk gaven, zaten de hoeveelheden in het vlees van deze koeien onder de wettelijke grens. Dat komt omdat dioxinen en dioxine-achtige PCB's via de melk worden uitgescheiden. Na de verplaatsing naar een schoner gebied daalden de berekende hoeveelheden. Bij volwassen runderen kwamen ze binnen anderhalve tot vier maanden tot onder de toegestane hoeveelheid. Bij kalveren duurde het langer dan zes maanden.

Dioxinen en dioxine-achtige PCB's zijn chemische stoffen die bij (vuil)verbranding zijn ontstaan. Ondanks de sterk gedaalde uitstoot in de laatste 25 jaar, komen ze nog steeds voor in Nederland. Bijvoorbeeld in gras, de bodem en rivierslib. Dioxinen kunnen schadelijk zijn voor het immuunsysteem, de ontwikkeling van de hersenen, en de voortplanting.

Kernwoorden: dioxinen, PCB's, runderen, uiterwaarden, overdrachtsmodellen, PBK-modellen, simulatie

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Summary

In 2020, elevated levels of the dioxins (polychlorinated dibenzo-p-dioxins and dibenzofurans, PCDD/Fs) and dioxin-like PCBs (DL-PCBs) were observed in muscle fat and liver of wild cattle grazing in floodplains in the Netherlands. The primary purpose of holding such cattle is nature management of the floodplains. To maintain the size and composition of the herds, animals are moved or slaughtered. Their meat is sold as 'wilderness meat' for human consumption.

In some instances the levels of dioxins and DL-PCBs in muscle fat were higher than the maximum level (ML) set in Commission Regulation (EC) No 1881/2006 on contaminants in foodstuffs. These elevated levels are most likely the result of the exposure of the wild cattle to dioxins and DL-PCBs via contaminated grass and adhering soil, or – for young calves – via contaminated milk. In particular, levels in soil are higher than those observed in regular pastures. A possible measure to lower the levels of these contaminants in cattle is to move them to cleaner areas with lower levels of dioxins and DL-PCBs in grass and soil, thereby reducing their exposure. Levels in the cattle will be reduced by elimination and further growth of the animals.

The Netherlands Food and Consumer Product Safety Authority (NVWA) has initiated research at Wageningen Food Safety Research (WFSR) to measure contamination levels of dioxins and DL-PCBs in tissues of cattle grazing in river floodplains, as well as in grass and soil from these sites.

NVWA, together with the Ministry of Health, Welfare and Sport (VWS) and the Ministry of Agriculture, Nature and Food Quality (LNV) requested RIVM and WFSR to model the transfer of dioxins and DL-PCBs from grass and adhering soil to meat of cattle grazing in floodplains.

Two research questions were formulated:

- 1) Can the dioxin and DL-PCB levels in meat from cattle grazing in floodplains be estimated during their life cycle, based on measurements in grass and soil?
- 2) Can it be estimated how long it takes to reduce the dioxin and DL-PCB levels in meat from cattle grazing in the floodplains to below the legal maximum levels after the animals are moved to a cleaner area with lower levels of dioxins and DL-PCBs in grass and soil?

Three physiologically based kinetic (PBK) models were developed to simulate the transfer of dioxins and DL-PCBs to meat (and more specifically muscle fat and liver) of cattle grazing in floodplains for: adult beef cattle (males and non-lactating cows), lactating cows and calves. These models were built based on a PBK model for dairy cattle that was previously developed by RIVM and WFSR. The 'Rode Geus' was chosen as model breed, since for this breed most analytical data, although still limited, were available. Using these PBK models, levels of dioxins and DL-PCBs (expressed in total toxic equivalents [total TEQ]) in meat of

cattle grazing on floodplains can be estimated based on analytical data from grass and soil.

Two exposure scenarios were used to simulate the transfer of dioxins and DL-PCBs: a worst case scenario and a realistic scenario. Both scenarios used analytical data from grass and soil samples from the floodplain of the river Waal in Beuningen. The worst case scenario was based on grass samples with high total TEQ levels due to disposition of contaminated sludge during a recent flooding. The realistic scenario was based on grass samples taken when the floodplains were not recently flooded. In both scenarios a seasonal variation in TEQ in grass was simulated.

The main results were as follows:

- Simulated total TEQ concentrations in muscle fat exceeded the ML of 4 pg/g fat in meat cattle, lactating cows and calves in both the worst case and the realistic scenario.
- In lactating cows, the concentrations in muscle fat are below the ML during part of the lactation period, due to the excretion of dioxins and DL-PCBs via the milk.
- Simulated total TEQ concentrations in the liver exceeded the ML of 0.5 pg/g wet weight in adult cattle in the worst case scenario and in calves in both the worst case and the realistic scenario.
- Simulated total TEQ concentrations in the liver were around or below the ML in adults cattle in the realistic scenario.
- The models predict that the period with the lowest total TEQ concentrations in muscle fat and liver is in the late summer to early autumn.

Moving cattle to cleaner areas was simulated by assuming that the daily total TEQ exposure dropped to a background level after the seasonal peak of the total TEQ concentration in meat is reached in the beginning of April. For adult cattle, it was assumed that the animals first grazed in the floodplains for 2.5 years. For calves, it was assumed that they were first exposed to dioxins and DL-PCBs solely via the milk for 6 months.

Following the realistic scenario, the time it takes for the total TEQ concentration to drop below the ML for muscle fat was 37 days, 24 days and more than 6 months, respectively, in beef cattle, lactating cows and calves. Following the worst-case scenario, it was 110 days, 76 days and more than 6 months, respectively, in beef cattle, lactating cows and calves. For the liver, this time was equal or shorter. When the animals would be moved at another time of the year when the total TEQ levels in muscle fat and liver are lower, the time until the levels drop below the ML will be shorter.

These simulations can be used as an indication for the best time of slaughter, but should be verified with actual measurements of dioxins and DL-PCBs in muscle fat and/or liver.

A first verification of the model showed that total TEQ concentrations measured in kidney fat and liver of three lactating cows that grazed in the floodplain in Beuningen were in good agreement with the model simulations of the worst case exposure scenario and close to the realistic

exposure scenario. It was assumed that the TEQ levels measured in kidney fat represent those in muscle fat when expressed on lipid weight. First results of blood fat analyses in male beef cattle showed a decrease in total TEQ of more than 90% over a period of 75 days when the animals were moved to a cleaner area. This gives a first indication that the anticipated and simulated decreases in total TEQ level in fat indeed occur. To further verify and optimize the models, more data on total TEQ levels in grass, soil and animal products are needed. Also, it is recommended to collect more data on the growth and the fat levels of the Rode Geus animals (both male and female), as well as seasonal variation of consumption of grass and soil, and changes over life-time. To extend the model to other breeds, information on other breeds should be collected as well.

At a later stage, the models could be further refined by considering possible differences in kinetics of the different congeners. In particular differences between dioxins and DL-PCBs could be of interest, given the relatively high contribution of DL-PCBs to the TEQ level in kidney fat compared to grass and soil.

1 Introduction

In 2020, elevated levels of dioxins (polychlorinated dibenzo-p-dioxins and dibenzofurans, PCDD/Fs) and dioxin-like PCBs (DL-PCBs) were observed in muscle fat and liver of wild cattle grazing in floodplains in the Netherlands (NVWA, personal communication). Wild cattle live in a herd consisting of calves, adult cows and bulls and graze in the floodplains throughout the year. The primary purpose of holding such cattle is nature management of the floodplains and not production of food of animal origin. To maintain the size of the herds, and to maintain a good balance between cows and bulls, animals are moved or slaughtered. Their meat is sold as 'wilderness meat' for human consumption.

In some instances the levels of dioxins and DL-PCBs in such wilderness meat were found to be higher than the maximum levels (MLs) set in Commission Regulation (EC) No 1881/2006 on contaminants in foodstuffs (EC 1881/2006; NVWA, personal communication). These elevated levels are most likely the result of the exposure of wild cattle to dioxins and DL-PCBs via contaminated grass and adhering soil (Traag et al., 2006). Young calves will initially be exposed mainly through contaminated milk via the mothers. In particular levels in soil are higher than those observed in regular pastures, most likely due to disposition of contaminated sludge after flooding of the floodplains. A possible measure to lower the levels of contaminants in animals is to move them to cleaner areas and thus reducing their exposure. Existing levels will be reduced by further growth of the animals and elimination of the contaminants.

The Netherlands Food and Consumer Product Safety Authority (NVWA) has initiated research to measure the contamination levels of dioxins and DL-PCBs in meat of cattle grazing in floodplains, as well as in grass and soil in the floodplains where the animals were foraging at the time of slaughter. This work is performed by Wageningen Food Safety Research (WFSR, 2021).

NVWA, together with the Ministry of Health, Welfare and Sport (VWS) and the Ministry of Agriculture, Nature and Food Quality (LNV) requested RIVM and WFSR to model the transfer of dioxins and DL-PCBs of grass and soil to meat of cattle grazing in floodplains.

Two research questions were formulated:

- 1) Can the dioxin and DL-PCB levels in meat from cattle grazing in floodplains be estimated during their life cycle, based on measurements in grass and soil?
- 2) Can it be estimated how long it takes to reduce the dioxin and DL-PCB levels in meat from cattle grazing in the floodplains to below the legal maximum levels after the animals are moved to a cleaner area with lower levels of dioxins and DL-PCBs in grass and soil?

For this purpose, three physiologically based kinetic (PBK) models were developed that simulate the transfer of the dioxins and DL-PCBs to muscle fat and liver of wild cattle grazing in floodplains for 1) adult beef cattle (bulls and non-lactating cows), 2) lactating cows and 3) calves. These models were built based on a PBK model for dairy cattle that was initially developed by Derks et al. (1993; 1994) and later modified for application in case of feed incidents (Hoogenboom et al., 2010) and for exposure during grazing (Traag et al., 2006; Zeilmaker et al., 2013). Dioxins and DL-PCBs accumulate in fatty tissues. The ML for these contaminants in meat is expressed on a lipid base. Also, an ML is set for the liver. Therefore, the focus of this research is on the levels in muscle fat and in the liver. Seasonal influences on dioxin and DL-PCB intake and body fat distribution of this type of cattle were taken into account in the modeling. Different breeds of cattle graze in the floodplains. In consultation with NVWA, VWS and LNV, one (generic) breed of cattle (the 'Rode Geus') was chosen as model breed. This is the breed for which most analytical data, although still limited, are available in the Netherlands.

This report describes the model simulations performed to answer the research questions. Detailed documentation of the models is published separately (Minnema et al., 2021).

2 Methods

2.1 General model overview

A schematic overview of the PBK models developed to estimate the level of dioxins and DL-PCBs is given in Figure 1.

Dioxins and DL-PCBs consist of a mixture of congeners that show the same toxicity. The potency of the different congeners varies greatly. Therefore, toxic equivalency factors (TEFs) are used to express their contribution in a mixture as an equivalent amount of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD, the most potent representative of the group) by multiplication of its concentration with the substance specific TEF. In this way, concentrations of dioxins and DL-PCBs can be expressed as total toxic equivalents (total TEQ).

For adult cattle (i.e., lactating cows and beef cattle), the intake of dioxins and DL-PCBs is modelled as the sum of total TEQ taken in from grass and from adhering soil. The intake of dioxins and DL-PCBs through the consumption of other feed materials such as leaves from other plants and small trees was not taken into account. For 0 to 6-months-old calves, milk was modelled as the sole contributor to the total TEQ intake, because (young) calves almost exclusively drink milk (Zeilmaker et al., 2013). For 6 to 12-months-old calves, the intake of total TEQ was modelled as the sum of TEQ taken in from grass and from adhering soil.

In the PBK models, it is assumed that after oral uptake, the compounds enter the gastro-intestinal (GI)-tract and are transported via the hepatic portal vein to the liver, instead of entering the systemic blood flow first. This is described previously in the models on which these models are built (Derks et al., 1993; Derks et al., 1994; Hoogenboom et al., 2010; Traag et al., 2006; Zeilmaker et al., 2013). After being taken up by the liver, the compounds can enter the systemic blood flow compartment, deposit in the liver or be excreted through hepatic clearance. The compounds in the systemic blood flow compartment can be distributed over the remaining compartments of the PBK model i.e., fat compartment, slowly perfused organs and richly perfused organs. In addition, in lactating cows, the compounds can be excreted from the systemic blood flow compartment through milk.

Lactating cows are assumed to primarily give milk in the spring and summer period (roughly April – September). For our simulation, a milk production of 7.5 L per day was assumed for a period of 6 months, starting in April. Therefore, the clearance through milk was only implemented for this period. When producing milk, cows have an increased cardiac output as explained by Minnema et al. (2021). This was also taken into account in the lactating cow model.

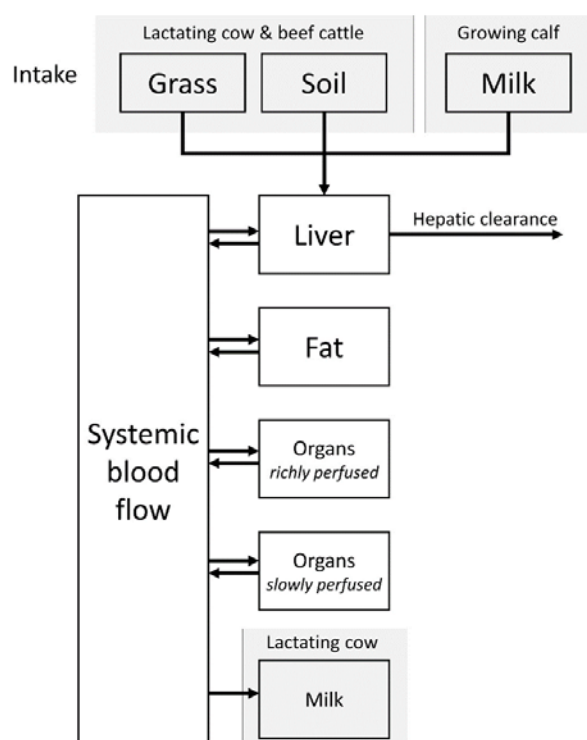


Figure 1 Schematic overview of the PBK model of the dioxins and DL-PCBs transfer routes in wild cattle living in a typical herd at the floodplains. Adult cattle (lactating cows and beef cattle) and 6 to 12-months-old calves are exposed to dioxins and DL-PCBs via grass and soil, whereas calves younger than 6 months take up these compounds through the milk. Excretion is solely performed through hepatic clearance, except for the lactating cows that also excrete dioxins and DL-PCBs through milk.

In adult cattle, the size of the fat compartment varies during the seasons. More specifically, cattle typically store fat in the summer when feed is abundant, and mobilize body fat in the winter when feed is more scarce and it will cost more energy to obtain the same feed intake for the herds, as also described by Minnema et al. (2021). In the model, this variation in body weight was implemented as a sinusoidal function with an amplitude of 26 kg (see Figure 2). This amplitude was derived from data reported by Hoch et al. (2005) that showed that Saler cows on a summer diet were approximately 52 kg heavier than Saler cows on a winter diet. It was assumed that the weight variation could be completely attributed to variation in the fat compartment. Thus, the fat compartment increases in size during spring/summer and decreases in late autumn/winter. Overall, the fat compartment is assumed to vary between 46 kg in the winter and 98 kg in the summer period, i.e. by more than a factor 2 (Figure 2b) in the lactating cow. This variation in fat is also present in beef cattle (i.e. 46-98 kg).

Since no such data were available for the Rode Geus, it was assumed that the seasonal weight variation is comparable between Saler cattle and Rode Geus cattle.

A similar assumption was made for the calves. Values for the weight growth model for calves aged 0-12 months were taken from Domínguez-Viveros et al. (2014) as described by Minnema et al. (2021) and shown in Figure 3. This was also based on Salers. Note that the

body weights for both calves and adult cattle referred to are the body weights minus the weights of the gastro-intestinal content. It was assumed that the weight of the gastro-intestinal content is a fixed fraction, i.e. 25% of the mean total life body weight as was done for the dairy cow by Derks et al. (1994) (Minnema et al., 2021).

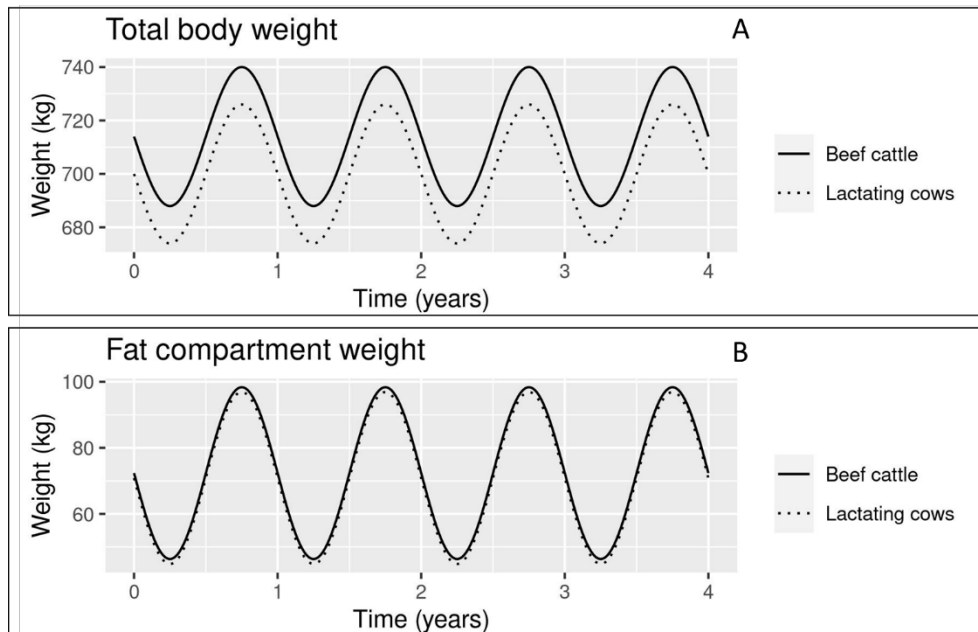


Figure 2 The total body weight variation (A) and the fat compartment weight variation (B) in beef cattle and lactating cows.

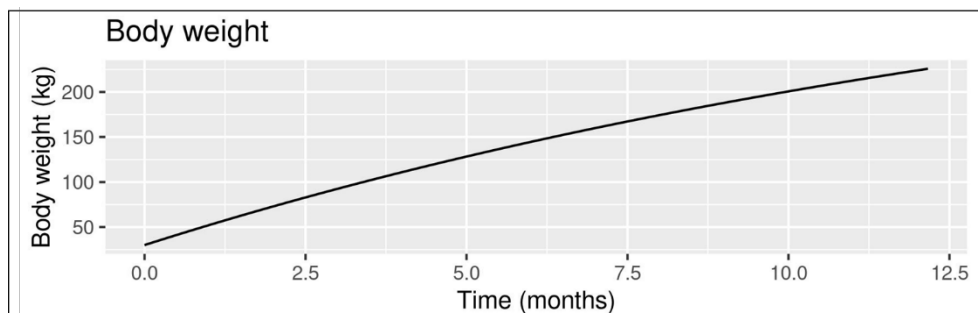


Figure 3 The growth curve for calves used in the simulation for calves aged 0-12 months.

The intake scenarios used in the PBK model simulations were a typical representation of a herd of wild cattle grazing at the floodplains year round. The intake scenarios used are explained below in chapter 3.2. Detailed documentation of the models developed is published separately (Minnema et al., 2021).

2.2 Sampling and analysis

Samples of grass and soil in the floodplains of Beuningen were collected in November 2020, February 2021 (just after the flooding) and April 2021. Grass and soil samples were collected at various distances from the river. In November 2020, also control samples across the river dike were collected. These were used to represent the background level of

TEQ in the Netherlands. In February 2021, three cows were slaughtered at the age of 1, 3 and 15 years. Samples of kidney fat and liver were collected at the slaughterhouse. All samples were analysed for dioxins DL-PCBs and (non-dioxin-like (NDL)-PCBs) at WFSR, using standard methods, accredited for animal derived products and feed. Levels are described in section 3.1. More details will be presented in another report (WFSR, 2021).

3 Exposure

3.1 Analytical data of grass, soil and kidney fat samples

The analytical data are shown in Table 1. TEQ levels in grass collected in November 2020 close to the river and close to the river dike were 0.7 and 0.9 ng TEQ/kg dry matter (dm), as compared to 0.3 ng TEQ/kg dm across the river dike. In February 2021, just after the flooding, grass samples collected at the bottom and halfway the river dike showed levels of 9.7 and 1.3 ng TEQ/kg dm. In samples collected mid-April 2021 halfway and near the river dike, levels were 0.4 and 0.2 ng TEQ/kg dm, respectively.

The levels in soil collected in November 2020 close to the river, halfway the flood plain and close to the dike were 7.1, 11.0 and 18.6 ng TEQ/kg dm, respectively. At the control site across the dyke it was 1.5 ng TEQ/kg dm. The level in soil collected after the flooding in February 2021 close to the river dike was 16.2 ng TEQ/kg dm.

In addition, TEQ levels in kidney fat and liver of three cows, slaughtered at the end of February 2021, were measured (see Table 1).

It should be pointed out that DL-PCBs contributed to a limited extent to the levels (18-44% in grass, 14-20% in soil). This was different from the observed levels in kidney fat (50 to 57% contribution from DL-PCBs) and suggests different kinetics or an additional exposure source (see Table 1). Additional sources could be leaves, barks, twigs and herbs.

Table 1 Concentrations of PCDD/Fs, DL-PCBs and the sum-TEQ in grass and soil and in kidney fat and liver of Rode Geus cows from floodplains near the Waal in Beuningen.

Sample	Sampling date	Levels ^{b, c}		
		PCDD/Fs (pg TEQ/g)	DL-PCBs (pg TEQ/g)	Sum TEQ (pg TEQ/g)
Grass (near river)	Nov. 2020	0.45/0.54	0.24/0.24	0.69/0.78
Grass (near dyke)	Nov. 2020	0.61/0.70	0.27/0.27	0.88/0.98
Grass (control)	Nov. 2020	0.21/0.37	0.12/0.12	0.33/0.49
Grass (foot of dyke) ^d	Feb. 2021	7.99/7.99	1.71/1.72	9.71/9.71
Grass (halfway dyke) ^d	Feb. 2021	0.95/1.07	0.24/0.24	1.18/1.31
Grass (middle flood plain)	Apr. 2021	0.05/0.22	0.16/0.17	0.21/0.39
Grass (near dyke)	Apr. 2021	0.03/0.17	0.06/0.07	0.09/0.23
Soil (near river)	Nov. 2020	5.9/5.9	1.2/1.2	7.1/7.1
Soil (middle flood plain)	Nov. 2020	9.0/9.0	2.0/2.0	11.0/11.0
Soil (near dyke)	Nov. 2020	14.8/14.8	3.7/3.7	18.6/18.6
Soil (control)	Nov. 2020	1.3/1.3	0.3/0.3	1.5/1.5
Soil (near dyke) ^d	Feb. 2021	13.9/14.0	2.2/2.2	16.1/16.2
Cow kidney fat (12-11-2019) ^a	Feb. 2021	5.5/5.5	7.3/7.3	12.8/12.8
Cow kidney fat (18-6-2005) ^a	Feb. 2021	6.5/6.5	6.5/6.5	12.9/12.9
Cow kidney fat (9-6-2017) ^a	Feb. 2021	5.1/5.1	6.2/6.2	11.2/11.2

Sample	Sampling date	Levels ^{b, c}		
		PCDD/Fs (pg TEQ/g)	DL-PCBs (pg TEQ/g)	Sum TEQ (pg TEQ/g)
Cow liver (12-11-2019) ^a	Feb. 2021	0.5/0.6	0.5/0.5	1.0/1.1
Cow liver (18-6-2005) ^a	Feb. 2021	1.3/1.3	0.7/0.7	2.0/2.0
Cow liver (9-6-2017) ^a	Feb. 2021	0.7/0.7	0.5/0.5	1.3/1.3

a Date of birth of the cows

b Lowerbound/upperbound levels, meaning that non-detected levels are assumed equal to zero or to the LOQ, respectively.

c Levels in kidney fat per gram fat, in grass and soil per gram 100% dry matter (grass recalculated from 88% dm)

d Samples collected just after the flooding of the floodplains in February

3.2 Exposure scenarios

Two exposure scenarios were used to simulate the transfer of dioxins and DL-PCBs, a realistic and a worst case scenario. The total TEQ concentrations in grass and soil applied in the model simulations were based on samples collected in the floodplains of the river Waal in Beuningen. The worst case scenario for adult cattle was based on grass samples collected in floodplains that were recently flooded and had high total TEQ levels due to disposition of contaminated sludge. The realistic case scenario was based on samples taken in floodplains that were not recently flooded. The worst case and realistic scenario for calves were based on the simulated mean total TEQ concentrations in milk from lactating cows exposed to a worst and realistic scenario, respectively. To simulate moving cattle to cleaner areas, it was assumed that the daily total TEQ exposure dropped to a background level following both exposure scenarios. An overview of all exposure scenarios and the minimal and maximal measured TEQ concentrations used for grass and soil and the average simulated TEQ concentration used for milk is shown in Table 2. A more detailed description for each cattle type is given below.

Table 2 Overview of exposure scenarios and the TEQ concentrations in grass, soil and milk used in these scenarios.

Wild cattle	Exposure scenario	Concentrations in grass (min-max)/soil (ng total TEQ/g dm) and milk (ng total TEQ/L)	Exposure period (days)
Beef cattle	Realistic ^a	Grass: 0.3 - 0.7 Soil: 16.2	820
	Worst case ^b	Grass: 0.3 - 9.7 Soil: 16.2	820
	Background level ^c	Grass: 0.3 - 0.5 Soil: 1.5	640
Lactating cows	Realistic ^a	Grass: 0.3 - 0.7 Soil: 16.2	820
	Worst case ^b	Grass: 0.3 - 9.7 Soil: 16.2	820
	Background level ^c	Grass: 0.3 - 0.5 Soil: 1.5	640

Wild cattle	Exposure scenario	Concentrations in grass (min-max)/soil (ng total TEQ/g dm) and milk (ng total TEQ/L)	Exposure period (days)
Calves 0-6 months	Realistic ^d	Milk: 0.19	182.5
	Worst case ^e	Milk: 0.57	182.5
Calves 6-12 months	Background level ^c	Grass: 0.3 – 0.5 Soil: 1.5	182.5

dm: 100% dry matter, min-max: minimal and maximal concentration in grass

^a Realistic scenario: Grass min: the average of the lowerbound/upperbound (LB/UB) TEQ levels in samples collected in middle flood plain in April 2021; Grass max: the average of the LB/UB TEQ levels in samples collected near river in Nov. 2020, Soil: the average of the LB/UB TEQ levels in samples collected near dyke in Feb. 2021

^b Worst-case scenario: Grass min: the average of the LB/UB TEQ levels in samples collected in middle flood plain in April. 2021; Grass max: the average of the LB/UB TEQ levels in samples collected at foot of dyke near river in Nov. 2020; Soil: the average of the LB/UB TEQ levels in samples collected near dyke in Feb. 2021

^c Background levels: Grass min: LB TEQ levels in samples from collected in cleaner area (control) in Nov. 2020; Grass max: UB TEQ levels in samples collected in cleaner area (control) in Nov. 2020; Soil: the average of the LB/UB TEQ levels in samples collected in samples collected in cleaner area (control) in Nov. 2020.

^d Realistic scenario calf: Average TEQ concentrations in the milk estimated by the model for lactating cows exposed to the realistic exposure scenario.

^e Worst-case scenario calf: Average TEQ concentrations in the milk estimated by the model for lactating cows exposed to the worst-case exposure scenario.

3.2.1 Adult beef cattle

For adult beef cattle, the intake of grass was assumed to be 15 kg dry matter/day. This intake was also used in a previous evaluation by RIVM and WFSR of dioxins in wild cattle (Zeilmaker et al., 2013). Similarly, adhering soil was assumed to be 4% of the total grass intake, which amounts to 0.6 kg soil intake per day. The absorption fraction of TEQ from grass was set to 0.25, whereas the absorption fraction of TEQ from soil was set to 0.43 (in concordance with Zeilmaker et al., 2013).

As described by Minnema et al. (2021) in detail, in the transfer models it was assumed that the maximum total TEQ concentration in grass would be reached just before the grass started to grow at the start of spring/summer (April 1st), and the minimum concentration would be reached just before the grass stopped growing at the start of autumn/winter (September 30th). This leads to a seasonal variation of TEQ intake via grass.

In the realistic scenario, data from samples collected when floodplains were not recently flooded were used. The maximum total TEQ concentration applied for grass was 0.7 ng/kg dry matter (from a sample collected in mid-November), and the minimum concentration applied was 0.3 ng/kg dry matter (from a sample collected in mid-April). There were no samples available that were collected at the end of summer / beginning of winter or at the end of winter / beginning of summer, but these measured concentrations suggest that in mid-April the grass was growing as is normal in spring/summer, compared to the grass sampled in mid-November that had stopped growing as it does in winter. The total TEQ concentration applied for soil was 16.2 ng/kg dry matter.

In the worst case scenario, the total TEQ concentrations in grass and soil were based on data from floodplains that were recently flooded. As a result the level in grass was strongly increased due to attached sludge, whereas the concentration in soil did not change. Here the maximum total TEQ concentration applied for grass was 9.7 ng/kg dry matter (sample collected in February), while the minimum concentration remained 0.3 ng/kg dry matter (sample collected in April). The total TEQ concentration applied for soil was again 16.2 ng/kg.

To simulate the effect of the transfer of the cattle to cleaner areas with lower concentrations of dioxins and DL-PCBs in grass and soil, the intake was changed to a background level at April 1st of the third simulation year. This was done for both the realistic and the worst case intake scenario. As background level a maximum total TEQ concentration in grass of 0.5 ng/kg dry matter, and a minimum concentration of 0.3 ng/kg dry matter were applied. The total TEQ concentration applied for soil was 1.5 ng/kg dry matter. Total TEQ intake from both grass and soil were based on control samples collected across the river dike in mid-November.

The resulting daily total TEQ intake in beef cattle and lactating cows via grass and soil following a realistic and worst-case scenario for 2.25 years (=820 days) followed by 1.75 years background exposure (=640 days) is depicted in Figure 4. These intake scenarios were used for the transfer of dioxins and DL-PCBs simulations in adult beef cattle and lactating cows. The daily total TEQ intake for cattle ranges from 5.3 to 41 ng TEQ/day for the worst case scenario and 5.3 to 6.8 ng TEQ/day for the realistic scenario. When exposed to background levels in grass and soil the daily intake ranges from 1.5 to 2.2 ng TEQ/day.

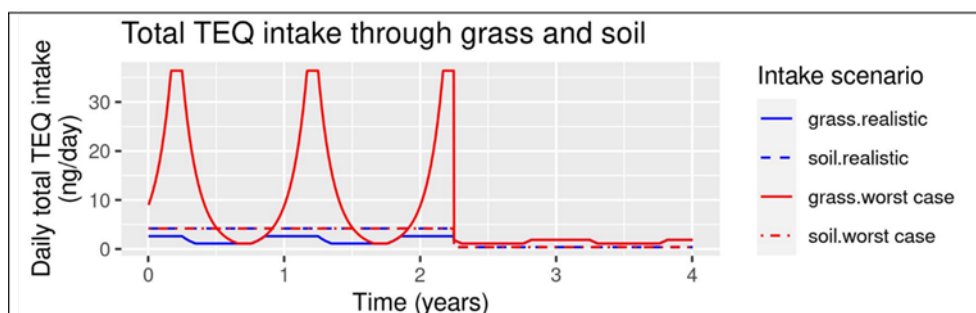


Figure 4 Daily total TEQ intake in adult beef cattle and lactating cows in the realistic intake scenario (blue) and the worst case intake scenario (red) through consumption of grass (line) and soil (dashed). After 2.75 years the daily total TEQ intake was calculated using background levels in grass and soil. The intake is corrected for the fraction absorbed.

3.2.2 Lactating cows

The exposure scenarios for lactating cows used for the simulations were similar to these for adult beef cattle described above (see Figure 4).

3.2.3 Calves

For the calf model, the TEQ intake for 0 to 6-months-old calves was assumed to be solely from drinking milk. Milk intake was modelled to be equal as the milk production of the lactating cattle (7.5 L/day) as

previously assumed by Zeilmaker et al. (2013). The mean total TEQ concentrations in the milk were simulated using the lactating cow model, resulting in total TEQ concentrations of 0.19 ng total TEQ/L in the realistic intake scenario and 0.57 ng total TEQ/L in the worst-case intake scenario (see Figure 8 in Chapter 4). Using an absorption fraction of 1.0 (assumed higher absorption via milk than feed) for total TEQ from milk, a daily intake of 1.4 ng total TEQ/day for the realistic scenario and of 4.3 ng total TEQ/day for the worst case scenario were calculated. These daily total TEQ intakes were used to simulate the resulting total TEQ concentration in the muscle fat of calves.

The effect of the transfer of calves to cleaner areas was simulated by assuming that the calves would replace their total TEQ intake via milk entirely by total TEQ intake via grass and soil based on background level contamination and absorption fraction of TEQ in grass and soil as was done for adult cattle. Except that the daily intake of grass by adults of 15 kg dry matter/day was scaled to the body weight of the calves. This resulted in a daily total TEQ exposure, starting around 0.4 ng TEQ/day at 6 months to 1.0 ng TEQ/day at 12 months.

The resulting total TEQ intake via grass and soil following a realistic and worst-case scenario for 6 months followed by 6 months background exposure is depicted in Figure 5. These intake scenarios were used for the transfer of dioxins and DL-PCBs simulation in calves. The background total TEQ intake through grass and soil, although increasing with age due to the gain in body weight and higher intake of grass and soil, is still clearly lower than intake through milk.

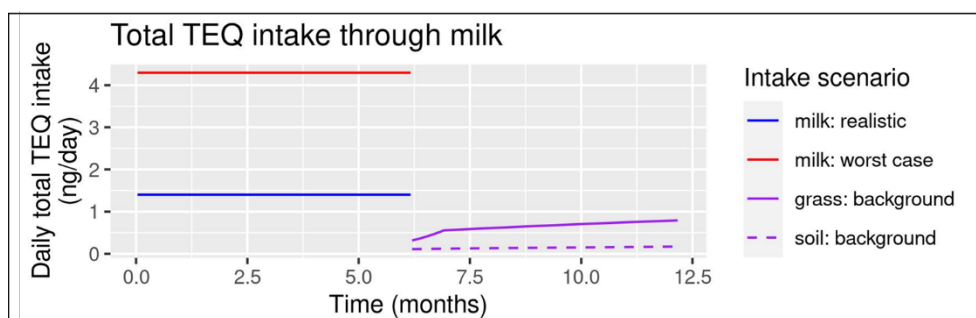


Figure 5 Daily total TEQ intake in calves in the realistic intake scenario (blue) and in the worst case intake scenario (red) through the consumption of milk. After 6 months the daily total TEQ intake was calculated using background levels in grass and soil. The intake is corrected for the fraction absorbed.

4 Simulation results for wild cattle grazing in floodplains

Simulations for the transfer of TEQs from grass and soil to animal products were performed with three transfer models for beef cattle, lactating cows and calves. Total TEQ concentrations in muscle fat and liver were simulated. These simulated total TEQ concentrations were compared to total TEQ concentrations measured in kidney fat and liver in lactating cows. It was assumed that the TEQ levels measured in kidney fat represent those in muscle fat when expressed on lipid weight. The results are described below.

For each model a simulation was run for 4 years, except for the calves for which a simulation of 1 year was run. For each model a realistic and worst-case intake scenario was taken into account. A worst-case scenario can occur after the floodplains are flooded thereby depositing contaminated river sludge on the grass.

Subsequently, for adult beef cattle and lactating cows, both intake scenarios were changed to background exposure at April 1st of the third simulation year, in order to simulate transfer of the animals to cleaner areas after long-term exposure to dioxins and DL-PCBs. For calves, both intake scenarios were switched after 6 months to background exposure. The background exposure was based on measurements in soil and grass taken from across the river dyke and represents the background level of dioxins and DL-PCBs contamination in the Netherlands.

4.1 Transfer of dioxins and DL-PCBs in adult beef cattle

Figure 6 shows the simulated total TEQ concentrations in muscle fat and liver following a realistic and worst-case exposure scenario for 2.25 years (=820 days) followed by 1.75 years background exposure (=640 days). A seasonal variation in total TEQ concentrations was seen for muscle fat and liver due to the seasonal variation in dioxin and DL-PCB intake (Figure 4) and body fat (Figure 2). As shown in figure 6A, the total TEQ concentrations in muscle fat varied between approximately 7.0 and 32 pg TEQ/g muscle fat for the worst-case scenario and were above the ML for dioxins and DL-PCBs of 4 pg TEQ/g fat for bovine meat and fat. In the realistic exposure scenario, the total TEQ concentration was just above the ML, the concentration in muscle fat ranged from approximately 4.4 to 7.7 pg TEQ/g muscle fat. As shown in figure 6B, the total TEQ level in the liver varied between approximately 0.5 and 2.1 pg TEQ/g liver in the worst-case scenario and was above the ML for dioxins and DL-PCBs of 0.5 pg TEQ/g liver. In the realistic scenario, the total TEQ level was between 0.3 and 0.5 pg TEQ/g liver and is just below the ML.

When it is simulated that the cattle is moved to cleaner areas in spring in the third year (April 1st), it took about 1.2 months (approximately 37 days) until the total TEQ concentration in muscle fat dropped below the ML of 4 pg/g fat (indicated by red horizontal dashed line) in the realistic scenario, whereas it took about 3.6 months (approximately 110 days) in the worst-case scenario (Figure 6A). For the total TEQ concentration in liver, to drop below the ML of 0.5 pg/g liver it took 0 days in the realistic scenario and about 2 months (approximately 65 days) in the worst-case scenario after moving to cleaner areas (Figure 6B).

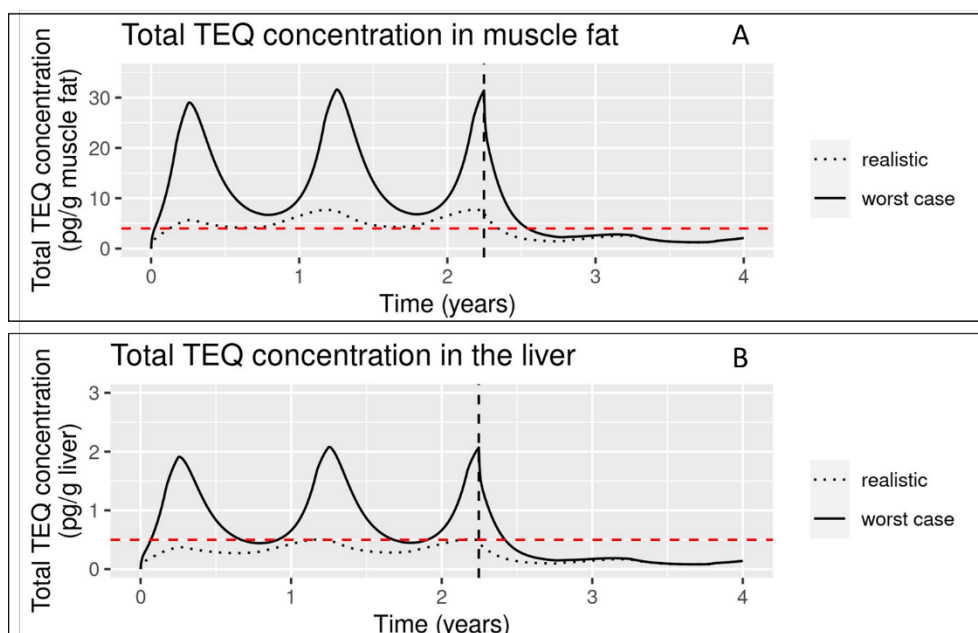


Figure 6 The simulated total TEQ concentration in muscle fat (A) and the liver (B) of adult beef cattle for the whole simulation duration (=4 years) for the realistic (black dotted line) and worst case intake scenario (solid black line). At $t=820$ days a move to cleaner areas was simulated as indicated by the black vertical dashed line). The ML for total TEQ in muscle fat and liver is indicated by the red horizontal dashed line.

4.2 Transfer of dioxins and DL-PCBs in lactating cows

Figure 7 shows a similar set of graphs as depicted in Figure 6, but for lactating cows that were living with their calves in the herd. In these simulations, dioxins and DL-PCBs were cleared by the liver, as well as via milk excretion (7.5 L milk/day). The lactation period in the model was set for 6 months a year (i.e. April 1st till October 1st). In Figure 8 the simulated concentrations in milk are shown for both the realistic and worst-case exposure scenario. The mean total TEQ concentrations in milk were 0.19 ng TEQ/L in the realistic scenario, and 0.57 ng TEQ/L in the worst-case scenario. These concentrations were used to simulate the resulting total TEQ concentration in the muscle fat of calves drinking this milk.

Similar to the adult beef cattle, the model simulations taking into account the seasonal variation in total TEQ intake and body fat variation resulted in a seasonal variation in the total TEQ concentration in muscle fat and liver of the cows. In the worst-case scenario, the concentrations in muscle fat varied from approximately 4.5 to 32 pg/g and exceeded the ML of 4.0 pg TEQ/g fat. In the realistic exposure scenario, the total TEQ concentration in muscle fat was around the ML, varying from 3.3 to approximately 7.7 pg TEQ/g muscle fat. In the liver, the concentrations varied between approximately 0.3 and 2.1 pg TEQ/g liver in the worst case scenario and between 0.2 and 0.5 pg TEQ/g liver in the realistic scenario. In the worst case scenario, the ML of 0.5 TEQ/g liver was exceeded, whereas in the realistic case scenario the values were just at or below the ML.

For three lactating cows that had grazed in the floodplains in Beuningen, slaughtered at the end of February, values in kidney fat and liver were measured (see paragraph 3.1). To see whether the model is close to actual measurements, a first verification of the model was done using these levels. The measured values (i.e. 11.2, 11.8 and 12.9 pg TEQ/g fat and 1.1, 2.0 and 1.3 pg TEQ/g liver) are shown as red dots in Figure 7A and 7B, respectively. It was assumed that the TEQ levels measured in kidney fat represent those in muscle fat when expressed on lipid weight. The dots appear to lie on the worst-case scenario line for both muscle fat (Figure 7A) and liver (Figure 7B) and are relatively close to the realistic exposure scenario line. Furthermore, the modelling suggests that the levels would have further increased with a peak at the end of March. However, additional data are needed to confirm this preliminary verification. Ideally, muscle fat and/or kidney fat and liver samples should be collected at different time points throughout the simulated period in combination with grass and soil samples.

When it is simulated that the cows are moved to cleaner areas in spring, it took about 24 days until the total TEQ concentration in muscle fat dropped below the ML of 4.0 pg/g fat (indicated by red horizontal dashed line) in the realistic scenario, whereas it would take about 76 days in the worst-case scenario (Figure 7A). For the total TEQ concentration in liver to drop below the ML of 0.5 pg/g liver, it took 0 days in the realistic scenario and about 46 days in the worst-case scenario after moving to cleaner areas (Figure 6B).

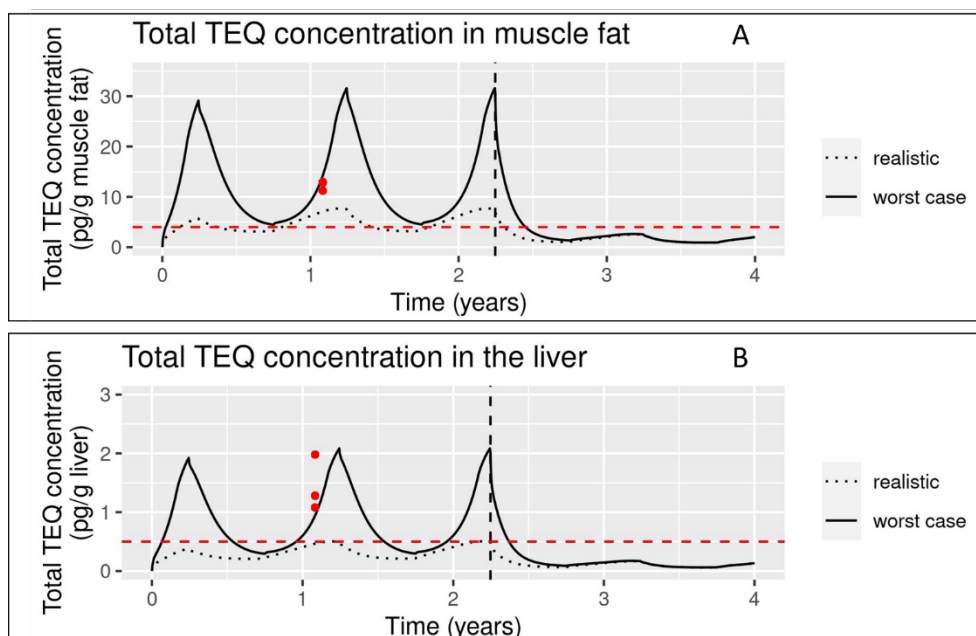


Figure 7 The simulated total TEQ concentration in muscle fat (A) and the liver (B) of lactating cows for the whole simulation duration (=4 years) for the realistic (black dashed line) and worst case intake scenario (solid black line). Lactation occurred in each year for six months, starting in April. Red dots represent measured values in kidney fat ($n=3$) (2 measurements overlap) and liver ($n=3$). At $t=820$ days a move to cleaner areas was simulated as indicated by the black vertical dashed line). The maximum level for total TEQ in muscle fat and liver is indicated by the red horizontal dashed line.

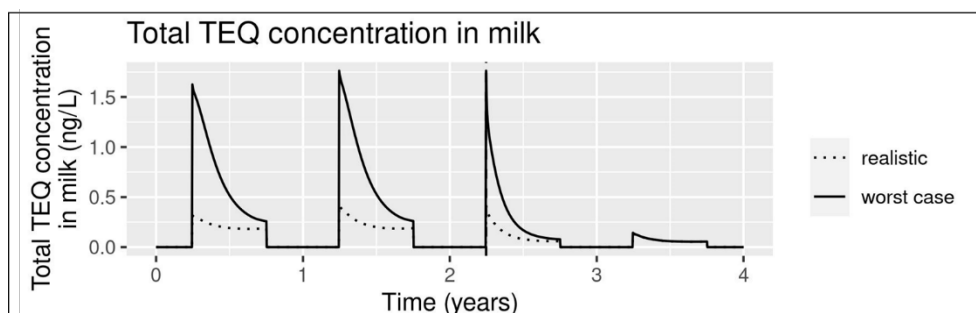


Figure 8 Simulated total TEQ concentration in milk produced by lactating cattle for the realistic (dashed line) and worst case intake scenario (solid line). At $t=820$ days a move to cleaner areas was simulated as indicated by the black vertical dashed line (black vertical dashed line overlaps with the start of the 3rd lactation period). Lactating periods were assumed to be from April 1st till October 1st (6 months).

4.3 Transfer of dioxins and DL-PCBs in calves

Figure 9 shows the simulated total TEQ concentration in muscle fat and liver using a realistic and worst-case exposure scenario in which calves were solely exposed via milk for 6 months, followed by 6 months background exposure via grass and soil. The model simulation resulted in a maximum total TEQ concentration around 26.8 pg TEQ/g muscle fat for the worst case scenario and 8.8 pg TEQ/g muscle fat for the realistic intake scenario (Figure 9A). This was above the ML of 4.0 pg/g fat. As seen in figure 9B, the maximum total TEQ concentration was around 1.8 pg TEQ/g liver for the worst case scenario and 0.6 pg TEQ/g liver for the realistic intake scenario. Both scenarios resulted in an exceedance of the ML of 0.5 pg TEQ/g fat in liver.

After switching to background exposure via consumption of solely grass and soil, it took more than 6 months until the total TEQ concentration in muscle fat dropped below the ML of 4.0 pg/g fat (indicated by red horizontal dashed line) in the realistic scenario and worst case scenario (Figure 9A). For the total TEQ concentration in liver to drop below the ML of 0.5 pg/g liver, it took about 7 days in the realistic scenario and more than 6 months in the worst-case scenario after moving to cleaner areas (Figure 6B).

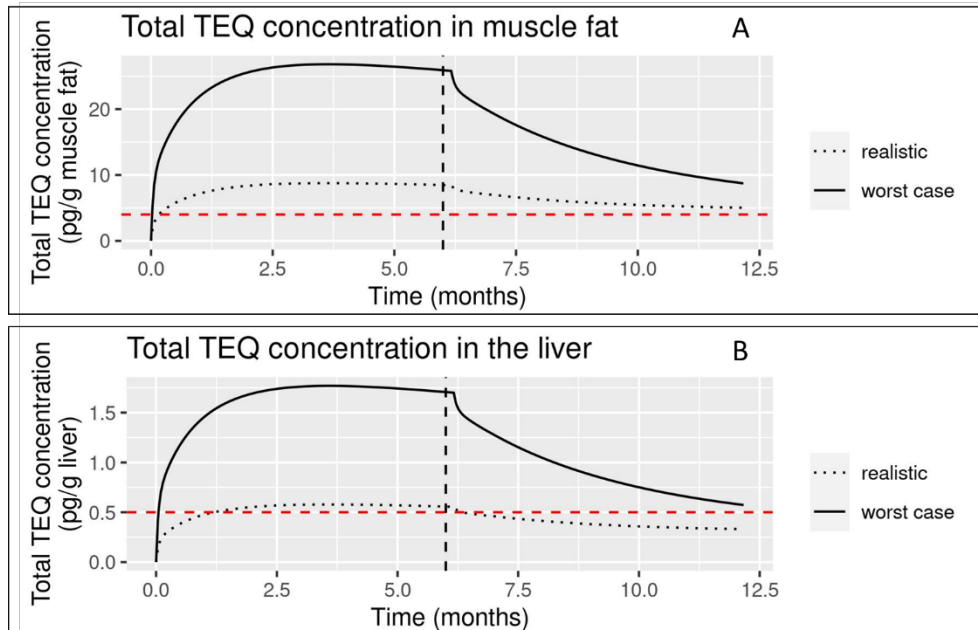


Figure 9 The simulated total TEQ concentration in muscle fat (A) and the liver (B) of calves for the whole simulation duration (=1 year, with exposure during 6 months via milk, followed by exposure to background levels via grass and soil) for the realistic (black dashed line) and worst case intake scenario (solid black line). The start of the last 6 months is indicated by the black vertical dashed line. The maximum level for total TEQ in muscle fat and liver is indicated by the red horizontal dashed line.

5 Discussion and conclusion

5.1 Discussion

5.1.1 *Model verification*

Three transfer (PBK) models are developed that can be used to estimate levels of dioxins and DL-PCBs in muscle fat and liver from wild cattle grazing in floodplains during their life cycle, based on measurements in soil and grass. The models can also be used to estimate how long it will take to reduce these dioxin and DL-PCB levels to levels below the maximum levels (MLs) after the animals are moved to a cleaner area with background levels of dioxins and DL-PCBs in grass and soil.

To verify whether the model simulations are close to the actual situation in wild cattle grazing in floodplains, a preliminary verification was performed using TEQ values in kidney fat and liver measured in three lactating cows that grazed in floodplains near the Waal in Beuningen (see paragraph 4.2, Figure 7). It was assumed that the TEQ levels measured in kidney fat represent those in muscle fat when expressed on lipid weight. The simulated total TEQ concentrations in muscle fat and liver in the worst case scenario closely resembled the measured TEQ concentrations, whereas the simulated TEQ concentrations in the realistic scenario resulted in a slight underestimation. This is a first indication that the lactating cow model provides adequate simulations of the TEQ concentration in muscle fat and liver. No measurements were available in milk from these cows for verification, since milk cannot be easily sampled in wild cows. No TEQ level measurements were available for calves and beef cattle from Beuningen.

To further verify the models, more data are needed and ideally muscle fat and/or kidney fat and liver samples should be collected at different time points from cattle grazing in the floodplains in combination with grass and soil samples of these floodplains, as well as from cattle moved to cleaner areas in combination with grass and soil samples from these areas. Data are currently collected for cattle that grazed in floodplains near Loevestein, including concentration data in blood fat. The first data in male beef cattle (n=5) showed a decrease in total TEQ in blood fat of more than 90% over a period of 75 days when the animals were moved to a cleaner area (WFSR, 2021). This gives a first indication that the anticipated and simulated decreases in total TEQ level in fat indeed occur. Whether total TEQ concentrations measured in blood fat can indeed be used as a biomonitoring tool for TEQ concentrations in kidney fat and meat fat needs to be investigated on the basis of data that are currently being collected.

5.1.2 *Simulations following exposure scenarios*

For adult cattle and calves, both the realistic and worst-case exposure scenario simulations resulted in TEQ concentrations in muscle fat exceeding the ML of 4 pg/g fat (EC 1881/2006). Also the simulated levels in liver exceeded the ML of 0.5 pg/g wet weight in the worst-case scenario for adult cattle and calves. In the realistic scenario, the values were below the ML, except for calves. The period with lowest TEQ

concentrations in muscle fat and liver appears to be in late summer to early autumn, although the total TEQ concentrations in muscle fat are still above the ML - except for lactating cows in the realistic scenario. For lactating cows, the concentrations in muscle fat are below the limit during part of the lactation period in this scenario, due to the additional excretion of dioxins and DL-PCBs via the milk. However, the lactation period did not affect the highest level in muscle fat at the start of the lactation period the following year.

A possible measure to lower the levels of contaminants in animals is by moving them to cleaner areas with lower contamination levels, thus reducing their exposure. The time it takes for the total TEQ concentration to drop below the ML for muscle fat was 37 days, 24 days and more than 6 months, respectively, in beef cattle, lactating cows and calves following the realistic exposure scenario. Following the worst-case scenario, it was 110 days, 76 days and more than 6 months, respectively, in beef cattle, lactating cows and calves. For the liver, this time was equal or shorter (see Table 3). More than 6 months follow-up could not be simulated for calves, since the growth curve (Figure 3) was only verified until 12 months.

Table 3 Time until total TEQ concentrations in muscle fat and liver were below the ML¹ in cattle moved to a cleaner area

Exposure scenario	Time until total TEQ concentrations in muscle fat or liver < ML ¹			
	Tissue	Adult beef cattle	Lactating cows	Calves (6-12 months)
Worst case	Muscle fat	110 days	76 days	> 6 months
	Liver	65 days	46 days	> 6 months
Realistic	Muscle fat	37 days	24 days	> 6 months
	Liver	NA	NA	7 days

¹ ML of PCDD/Fs and DL-PCBs is 4.0 pg TEQ/g fat (muscle fat) or 0.5 pg TEQ/g wet weight (liver) (EC 1881/2006)

NA: Not applicable, because TEQ concentrations were below the ML before the cattle was moved to a cleaner area.

In these simulations, the animals are moved to cleaner areas at the seasonal peak of total TEQ concentrations in meat. When the animals would be moved during another time of the year when the total TEQ levels are lower, the time it takes for TEQ concentrations to drop below the ML will be shorter.

These simulated periods can be used as an indication for the best time of slaughter, but should be verified with actual measurements of dioxins and DL-PCBs in muscle fat or liver.

In the current models, growth was only taken into account for the calves, and the adult cows were only assumed to vary in body weight due to seasonal variation in bodyfat. However, since TEQ levels might be reduced by further growth of the adult animals and in particular by the increase in the fat compartment as well, this might result in an overestimation of the total TEQ concentrations and of the duration until total TEQ levels are below the ML. The reduction in the levels shown in adult animals as simulated by the models is primarily a result of the

seasonal increase in fat levels (thus reducing the concentrations in fat) as well as an active elimination of the contaminants via the liver and for lactating cows the excretion via milk.

5.1.3 *Different breeds and livestock cattle*

The current models are developed using the Rode Geus as a model breed. Due to lacking physiological data for the Rode Geus, data from Saler cattle were used assuming growth and physiological parameters are similar between the two breeds. The Rode Geus is a crossbreed between Saler and Brandrode cattle. Other wild species also graze on the floodplains in the Netherlands, such as Highlander, Galloways and Tauros herds. To extend the current models to other breeds, it needs to be studied whether adjustments are needed due to possible differences in body weight, fat content, growth, grazing behaviour and energy requirements. Highlanders and Galloways are e.g. known to contain more body fat (WFSR, personal communication).

Besides wild cattle, also beef cattle from livestock farms can graze on the floodplains during a large part of the year. It is likely that beef cattle from regular farms only graze on the floodplains when grass is in abundance, i.e. (part of) spring, summer and autumn periods. It is likely that the exposure of these animals to dioxins and DL-PCBs in winter is lower compared to wild cattle in the floodplains, because they will then receive cleaner feed in the stables. In addition, next to differences in husbandry of livestock cattle compared to wild cattle, there might be differences in physiology, such as the size of the fat compartment and the age of the animals when sent to slaughter, as also mentioned by Lake et al. (2014). Therefore, the current set of models cannot be applied to this type of cattle. However, in general, lower total TEQ levels are expected in meat of cattle of regular livestock farms since their yearly TEQ intake is generally lower and they have a similar or bigger fat compartment than the wild cattle.

5.1.4 *Exposure scenarios*

To improve the model simulations, the exposure scenario could be improved based on additional measurements of total TEQ concentrations in grass and soil, also at low levels within relevant grazing areas, preferably at different timepoints during a year. Also, more information on the cattle's grazing behaviour such as grass and soil intake and consumption of other feed sources such as leaves, barks, twigs and herbs could be used for further refinement of all three PBK models. For Highlanders it is estimated that these other feed sources account for 30% of the total dry matter intake, but no information about seasonal variations in the consumption of these additional sources was described (Ma et al., 2001). One might argue that in the winter period, when grass is more scarce, relatively more other materials might be consumed. Yet, no TEQ concentrations were available for these feed materials. These were therefore not taken into account in the exposure scenarios.

Next to type of feed intake, the amount of grass and/or soil intake might also change during the season. In the models, it was assumed that both soil and grass intake are similar in all seasons. Due to the growth of grass in spring and summer, the concentration of total TEQ in grass varies over the year (as described by Minnema et al. (2021)). It could be reasoned

that in autumn and winter, grass will be shorter and relatively more soil might be consumed by cows whereas more grass will be consumed during summer months when more is available. This may also depend on the weather/flooding conditions. But, data are lacking to estimate the magnitude of this variation and this was therefore not taken into account. In the current simulation, we did include the seasonal influence on total TEQ concentration in grass due to growth of the grass. TEQ concentrations in soil are generally relatively constant, also after a period of flooding (see Table 1). This is in contrast to grass, where high amounts of sediment may remain on the vegetation after the flooding. This sedimentation on grass will gradually disappear due to consumption and/or precipitation. Other seasonal factors such as heavy precipitation were not taken into account.

This report focused on dioxins and DL-PCBs levels in cattle and in grass/soil. Yet, a number of other contaminants such as per- and polyfluoroalkyl substances (PFASs) are being analysed in the tissues of animals, and when elevated, also in grass, soil and water (WFSR, 2021). For these contaminants, transfer modelling could be performed in the future.

5.2 Conclusion

Levels of dioxins and DL-PCBs (total TEQ) in meat of wild cattle grazing in floodplains can be estimated based on levels measured in grass and soil using the three developed transfer models for dioxins and DL-PCBs in beef cattle, lactating cows and calves. Simulated concentrations in muscle fat and liver were above the ML in a worst case scenario based on concentrations in grass samples collected after the floodplains were flooded. Concentrations in muscle fat were also above the ML in a realistic scenario based on grass levels collected when the floodplains were not recently flooded, except for lactating cows where the total TEQ levels drop below the ML during part of the lactation period. Cattle intended for slaughter can be moved to a less contaminated area to reduce dioxin and DL-PCB levels in muscle fat and liver, as shown by simulations in which the animals were exposed to background levels after being exposed to contaminated grass and soil, or milk for a long period. The simulated times it takes until TEQ concentrations drop below the ML can be used as an indication for the optimal time to slaughter, but should be verified with actual measurements of dioxins and DL-PCBs in meat.

5.3 Recommendations

Currently, additional data are collected, such as blood and tissue levels in animals transferred to cleaner areas, as well as the contamination levels in grass and soil of these areas. Also more information is being collected on the characteristics of the wild cattle in the floodplains. It is recommended to use these data to verify and further improve the beef cattle, lactating cow, and calve models and the exposure scenarios.

To further optimize the models, it is recommended to collect more data on the growth and fat level of animals (both male and female), as well as on seasonal variation of consumption of grass and soil, and changes over life-time. Ideally, these data are collected for different breeds to

make the models more broadly applicable for wild cattle in the floodplains.

At a later stage, the models could be further refined by considering possible differences in kinetics of the different congeners. In particular differences between relevant dioxins and DL-PCBs could be of interest, given the relatively high contribution of DL-PCBs to the TEQ level in kidney fat compared to grass and soil as mentioned in chapter 3.1.

6 Acknowledgements

Annick van den Brand (RIVM), Ans Punt (WFSR) and Leonie Lautz (WFSR) are acknowledged for critical review of the report.

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