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Transgenerational mass balance and tissue distribution of PCBs and PCDD/Fs from grass silage and soil into cow-calf continuum

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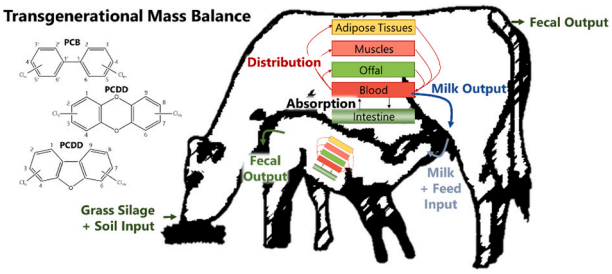
HIGHLIGHTS

- Increasing PCB lipophilicity reduced the absorption rate in cows but not in calves.
- Compared to POPs from grass silage, POPs from soil were less absorbed by cows.
- Calf exposure to POPs occurred more via milk suckling than prenatal exposure.
- Young calves (3 mo) had lower estimated metabolism rates than 10 mo calves and cows.
- IPCBs are more concentrated in liver, kidneys and muscles than in adipose tissue.

GRAPHICAL ABSTRACT



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ABSTRACT

Grass-based suckling beef-derived foods occasionally exceed regulatory levels for polychlorinated biphenyls (PCBs) and dibenzo-*p*-dioxins/dibenzofurans (PCDD/Fs). Ensuring chemical safety requires understanding the cow-calf transgenerational PCB and PCDD/F fate. The current focus was on dairy cows, omitting transgenerational fate and suckling beef-related physiological effects. This study aimed to investigate PCB and PCDD/F absorption, distribution, metabolism, and excretion within 12 Simmental cows (six primiparous/six multiparous) and 12 calves fed with the milk of their respective mothers for 109 days *prepartum* until 288 days in milk (DIM), i.e., slaughter time. Eight cows were exposed to a grass silage-soil mixture. Four were decontaminated after DIM164 by receiving uncontaminated grass silage, which four control cows received. An input-output balance during gestation and lactation was computed from PCB, PCDD/F, and lipid inputs (solid feed/milk intakes),

Abbreviations: ADME, Absorption, distribution, metabolization, excretion; AE, Assimilation efficiency; AR, Absorption rate; BW, Body weight; Ctl, Control; Dec, Decontaminated; DIM, Days in milk; dlPCB, Dioxin-like polychlorinated biphenyl; DM, Dry matter; DMI, Dry matter intake; EU, European Union; Exp, Exposed; GC-HRMS, Gas chromatograph-high resolution mass spectrometer; iPCB, Indicator polychlorinated biphenyl; K_{OW}, n-octanol-water parition coefficient; LT, *Longissimus thoracis*muscle; MS, Mass spectrometer; PBTK model, Physiologically-based toxicokinetic model; PCB, Polychlorinated biphenyl; PCDD/F, Polychlorinated dibenzo-*p*-dioxin/dibenzofuran; POP, Persistent organic pollutant; *RA*, *Rectus abdominis* muscle; TEQ, Toxic equivalent.

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outputs (fecal/milk excretions), and body storage (initial/final burdens). At slaughter, PCB and PCDD/F tissue distribution, and lipid allometry were linked. Apparent PCB and PCDD/F absorption rates and metabolized fractions decreased with increasing chlorination. In calves, PCB absorption showed no effect due to chlorination (steady range: 71–87%). High-chlorinated PCB and PCDD/F absorption rates decreased when provided through soil. Cows excreted PCBs and PCDD/Fs via feces (50% relative to input) and milk (9%) and accumulated only 5% in their body, whereas calves accumulated the largest fraction of the total input in their bodies (44%). Cow physiology affected accumulation and excretion, as in primiparous cows, net body burden and milk assimilation efficiencies were higher and lower, respectively, than in multiparous. Liver-specific enrichment was observed in cows and calves (7.0- and 3.2-fold iPCB and dlPCB + PCDD/F TEQ, compared to empty body-based lipid concentrations), whereas iPCBs were also enriched in kidneys (3.1-fold) and muscles (1.5-fold). Consequently, adipose concentrations did not perfectly represent most edible beef tissues. This highlights the essence of integrating the interplay between physicochemical pollutant properties and animal physiology in transgenerational transfer assessments of PCBs and PCDD/Fs.

1. Introduction

Polychlorinated biphenyls (PCBs) and dibenzo-*p*-dioxins/dibenzofurans (PCDD/Fs) are persistent organic pollutants (POPs) (EFSA, 2018; Stockholm Convention, 2022). Their easily dispersive, lipophilic, and persistent features explain their bioaccumulation in the environment and feed of livestock, with consequences for the chemical food safety of animal-derived products (EFSA, 2018). In addition to accidental point sources that form exposure pathways to livestock (e.g., accidently contaminated feedstuffs or old PCB-contaminated building materials), there are seemingly non-figurative and thus difficult to identify diffusive sources (Weber et al., 2018). Those result, e.g., from POP atmospheric emissions and depositions onto plants and topsoil, where they accumulate and persist, thereby exposing livestock – primarily those from grass-based husbandries (Schulz et al., 2005; Weber et al., 2019). Extensive suckling beef systems based on grass feeding (pasture, hay, and silage) have been found to occasionally exceed meat maximum regulatory levels (BAG, 2012; Weber et al., 2015; Zennegg, 2018). Bovine may involuntarily ingest 3% [dry matter (DM) intake basis of total diet] or more soil at pasture (Collas et al., 2019; Healy, 1968; Jurjanz et al., 2012), while milk suckling leads to cow-calf transgenerational bioaccumulation of POPs (Driesen et al., 2022; Hirako, 2008).

To ensure the chemical food safety and sustainability of grass-based suckling beef production systems, it is necessary to understand the fate of PCBs and PCDD/Fs that originate from diffusive grass silage and soil sources in transgenerational transfer settings. Transfer results from a series of toxicokinetic steps, including absorption, distribution, metabolization, and excretion (ADME) (Amutova et al., 2020). Only a few attempts at quantitative ADME assessments of dairy cows and non-lactating ewes have previously been reported (Kierkegaard et al., 2009; Lerch et al., 2020; McLachlan, 1993). Thus far, studies examining POP fate in suckling cow-calf pairs are scarce (Driesen et al., 2022; Hirako, 2008). Thanks to these first reports, PCB and PCDD/F kinetics in key distribution (blood and adipose tissue) and excretion (milk) compartments were described. Especially Driesen et al. (2022), demonstrated that 'global' oral intake to milk transfer rates and concentration kinetics in blood, adipose tissue, and milk depend not only on pollutant congener physicochemical properties, but also on animal physiological features (reproductive parity/milk yield, and lactation and growth stages). The need to decipher these respective effects on absorption and excretion (milk and feces) by considering not only concentrations but also mass dynamics was highlighted. For such an approach, a mass balance, as previously described by McLachlan (1993), from which the metabolized fraction can be estimated is needed. Moreover, deciphering the influence of POP properties and animal physiology on tissue distribution would not only be of great merit for toxicokinetic insights (i.e., the key roles of the liver and adipose tissue in metabolism and storage, respectively), but also for edible food product safety – i.e., muscles and intermuscular adipose tissue (meat), as well as liver and kidneys (offal). To the best of our knowledge, neither the mass balance nor tissue distribution of legally regulated PCB and PCDD/F congeners and the effects of physiology have been investigated in a transgenerational cow-calf setting.

The aims of this present study were (i) to set up a complete mass balance and (ii) to explore the tissue distribution of PCBs and PCDD/Fs from grass silage and soil in cows and their calves during gestation (109 d *prepartum*) and the entire lactation period [288 days in milk (DIM)]. Novelty relies on the fine quantification of PCB and PCDD/F ADME in cows and calves and understanding their link with physiological determinants; therefore, the focus was on body lipid dynamics.

2. Materials and methods

2.1. Animals and diets

The cattle experiment (n°2018_08_FR) was described in detail by Driesen et al. (2022). In brief, 12 pregnant Simmental bovine females (six primiparous and six multiparous) and their calves underwent an experimental period that lasted 109 ± 11.5 d *prepartum* until 288 ± 4.5 DIM (Fig. 1). The animals were divided into four control and eight exposed treatments based on parity and physiological traits. The exposed animals were further subdivided after DIM164 ± 4.4 to investigate decontamination kinetics in four cow-calf pairs. The control and decontaminated cows (after DIM164) were fed daily with grass silage only (low background POP levels), whereas the exposed group received grass silage mixed with 2.5% (DM basis) environmentally POP-loaded soil.

After calving, the cow-calf pairs were separated, and the calves were fed twice daily with the milk of their respective mothers. Additionally, calves received uncontaminated hay and, after three months of age, a concentrate feedstuff. Two calves died suddenly at three months of age due to rumen milk drinking (in the results, these calves are referred to as the 3 month-old exposed treatment group). At DIM288, the remaining cows and calves were slaughtered.

2.2. Measurements and sampling

2.2.1. Feed intake and milk and fecal excretion

For solid feed intake quantification, individual DM intake (DMI) was measured 4 d week^{-1} based on fresh matter intake (weighing of offered and refused) and bi-weekly DM content determination (60 °C, 20 h, followed by 105 °C, 3 h) of the offered feed. Grass silage, grass silage-soil mixture, hay, mineral, salt, and concentrate pools were composited per lot for chemical and POP analyses from dry subsamples (50 °C, 72 h), which were collected weekly or bi-weekly (for grass silage and grass silage-soil mixture) during the experiment. For the quantification of cow's milk excretion and calf milk intake, individual milk yield and intake were recorded at each milking by weighing the offered (i.e., the milk yield) and calf refusals. Individual milk pools representing the DIM1-164 (including the colostrum period) and DIM165-288 periods (only DIM1-288 pool for the control treatment, Fig. 1) were composited

for POP analyses from milk subsamples collected bi-weekly over two successive milking, proportionally to milk yield (i.e., cow milk excretion) or calf milk intake. For the quantification of cow and calf fecal excretion, the same pool periods as for milk, as well as one for the *prepartum* period for cows, were composited from bi-weekly individual dry subsamples (50 °C, 72 h) of feces collected straight from the rectum (Fig. 1). Pools were composited proportionally to DMI and estimated digestibility [based on ash, crude protein, and crude fiber content in feedstuffs (Driesen et al., 2022) using Prev'Alim software (INRA, 2006)]. Fecal fluxes of DM were further estimated using DMI and acid-insoluble ashes as indigestible marker measured in both diets and feces (Van Keulen and Young, 1977).

2.2.2. Postmortem tissue sampling and body burden

Detailed slaughter procedures and *postmortem* sampling were described by Xavier et al. (2022) (see **SI Section 1** for additional details). In brief, blood samples for POP analyses (600 g) were collected at exsanguination and clot at ambient temperature before centrifugation within 4 h (3,000 *g*, 15 min, 20 °C). The resulting blood serum was collected in glass bottles and stored at −20 °C.

After chilling carcass and non-carcass compartments for 24 h at 4 °C, tissue samples were collected in aluminum plates and stored at −20 °C, pending chemical and POP analyses (2–3 aliquots of 50–330 g each). From the right half-carcass, the *rectus abdominis* (*RA*) and *longissimus thoracis* (*LT*) muscles, and from the left 11th rib the intermuscular adipose tissue were fully dissected and weighed before being cut into small pieces and sampled. Perirenal and pericaudal subcutaneous adipose tissue, liver, and kidney were fully collected and weighed before being sampled after grinding and mixing using a mincing-cutter device. The entire left half-carcass (including the 11th rib) and rest of the empty body (full body minus digesta, urine, exsanguinated blood, and carcass) were further frozen at −20 °C before serial grinding, homogenizing, refreezing, and mincing procedures, followed by sampling of the homogenate (Xavier et al., 2022).

2.3. Chemical and POP analyses

The various chemical analyses performed were reported by Xavier et al. (2022) and Driesen et al. (2022). Feedstuffs, feces, muscles, liver, and kidneys were oven-dried (50 °C, 72 h) and ground (Grindomix GM200, Retsch, Haan, DE) before DM (105 °C, 3 h), and for feedstuffs and feces, lipids (ISO 6492:1999, petroleum ether extraction with a Büchi Speed Extractor E−916, Flawil, CH), and acid-insoluble ashes (Van Keulen and Young, 1977) were determined.

For PCB and PCDD/F analyses, previously reported analytical methods were followed (Driesen et al., 2022) (see SI Section 2 for a detailed account of the procedure and quality assurance). Briefly, dried and ground feedstuffs, feces, muscles, liver, and kidneys were Soxhlet extracted. Soxhlet extraction was also applied to the thawed samples of adipose tissues, carcass, and rest of the empty body (fresh matter basis). Blood serum and milk were liquid-liquid extracted. For animal samples, lipid content was determined gravimetrically, followed by spiking $^{13}C_{12}$-labeled indicator PCB (iPCB), dioxin-like PCB (dlPCB), and PCDD/F internal standards. Silica, alumina, and carbon column chromatography clean-up were used for purification. For the quantitative analyses of PCBs, a Q-Exactive Orbitrap GC-HRMS (ThermoFisher Scientific, MA, USA) was used, whereas PCDD/Fs were analyzed using an APGC-Xevo TQ-XS Triple Quadrupole MS (Waters, SM, USA). The toxic equivalent (TEQ) levels presented are based on the lower-bound (not detected = 0) WHO-toxic equivalency factors from 2005 (Van den Berg et al., 2006), except for the initial burden (i.e., initial POP mass stored in the empty body) calculations, where a medium-bound was used.

2.4. Calculations and statistical analyses

It was assumed that oral intake (i.e., of grass silage, soil, hay, concentrate, minerals, salts, and milk) was the only relevant exposure route (McLachlan, 1993), that feces and milk were the main excretion routes, and that the formation of parent compounds within cows or calves was negligible for hydrophobic and non-volatile PCBs and PCDD/Fs. Such assumptions resulted in the following equations (Kierkegaard et al., 2009; Lerch et al., 2020):

$$\text{Metabolism (ng)} = (\text{initial body burden} + \text{oral intake}) - (\text{final body burden} + \text{calf body burden}_{\text{parturition}} + \text{fecal excretion} + \text{milk excretion}), \quad (1)$$

$$\text{Absorption rate (AR; \%)} = (\text{oral intake} - \text{fecal excretion})/\text{oral intake} \times 100, \quad (2)$$

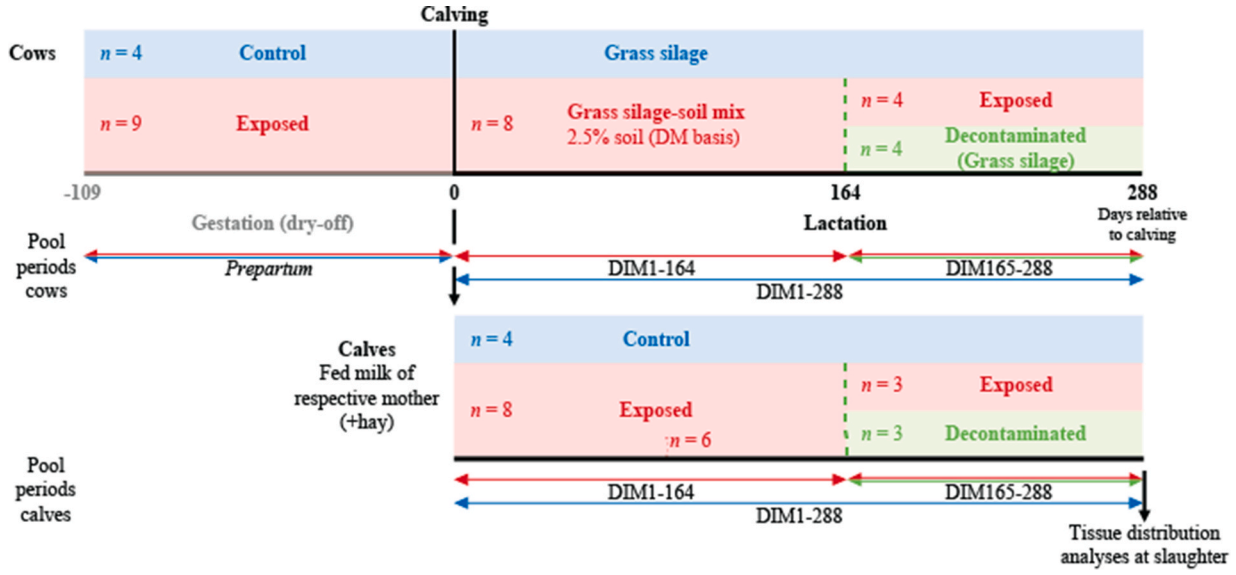

Fig. 1. Overview of the experimental design. Each treatment group equally consisted of primi- and multiparous cows, except for the gestation period, during which one additional primiparous exposed cow was present, and after days in milk (DIM) 89 for the calves with one exposed and one decontaminated primi-calf only, due to the early deaths at three months of age. Periods for the milk and fecal pool samples are indicated with arrows in the color of the corresponding treatment group for cows and calves. Samples for tissue distribution analyses were collected at slaughter (DIM288). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

$$\text{Assimilation efficiency (AE; \%)} = [(\text{final body burden} - \text{initial body burden}) + \text{milk excretion} + \text{calf body burden}_{\text{parturition}}]/\text{oral intake} \times 100, \quad (3)$$

where the calf body burden$_{\text{parturition}}$ and milk excretion terms are only specified for cows, whereas oral intake includes both solid feed and milk for calves.

The computation of final body burden of POPs in cows and calves is detailed in the footnote of Tables S3–4. The initial body burden in cows was estimated from the subcutaneous adipose tissue concentrations at the start of the experimental period (109 ± 11.5 d *prepartum*) multiplied by the estimated initial empty body lipid mass (detailed in footnote of Table S3). For calves at parturition, the adipose tissue DIM1 concentration of the respective mother cow was multiplied by the calves' body lipid mass at calving, assuming 4.5% lipids in the total body (Robelin, 1986). This estimation seemed appropriate, as the adipose tissue POP concentrations in a 7-day-old exposed calf accorded with those seen in exposed cows at DIM1 (Driesen et al., 2022).

The difference between AR and AE, resulted in the proportion of ingested POP that was not recorded at the end of the experiment and might therefore be attributed to the metabolized (i.e., biotransformed) fraction.

To explore the physiological determinants of lipophilic POP tissue distribution through body lipid accretion during the second half of lactation (cows) or growth (calves), allometric equations for lipid weights belonging to the three adipose tissue depots and the two muscles were calculated according to Atti and Bocquier (1999):

$$\ln(\text{adipose tissue or muscle lipids weight, kg}) = a \times \ln(\text{empty body lipids weight, kg}) - b, \quad (4)$$

where ln is the natural logarithm, and the slope, a, is the allometric coefficient.

Tissue distributions of PCBs and PCDD/Fs were analyzed by ANOVA using the MIXED procedure for repeated measurements of SAS (version 9.4, SAS Institute, Inc, NC, USA). For cows, concentrations of PCBs and PCDD/Fs on a total lipid basis in all tissues measured at slaughter were analyzed using a model that included dietary treatment (control, exposed, or decontaminated), tissue, parity (primi- or multiparous), and their interactions as fixed effects, cow as a random effect, and tissue as a repeated statement. For calves, the same model was used, but it included the 10-month-old control, 3-month-old exposed, 10-month-old exposed, and 10-month-old decontaminated groups as modalities for the dietary treatment effect and excluded the parity effect. The model used to analyze the AR and AE of each congener included dietary treatment, parity (only for cows), and their interaction as a fixed effect, and animal as a random effect. Logarithmic transformation was applied when needed to comply with the assumptions of normality, homoscedasticity, and linearity of residuals. Arithmetic means and standard errors reported were from the untransformed data, whereas *P*-values reflected transformed statistical analyses. The significance level was set at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$.

3. Results

3.1. Cow and calf lipid dynamics

Physiological traits, including feed intake, body weight (BW), body fatness, and milk and fecal excretion are listed in Tables S1–2. During the *prepartum* and entire lactation periods, lipids were lower in primiparous cows for intake (1.3-fold), calving excretion (1.2-fold), milk excretion (1.7-fold), and fecal excretion (1.3-fold) than in multiparous cows (multi: 190, 2.1, 182, and 73 kg lipids, respectively; Table S3). Conversely, the body lipid mass increase from DIM-109 to 288 was 3.6-fold higher in primiparous cows than in multiparous cows. In 10-month-old calves, total lipid intake (solid feed and milk) until DIM288 was 128 kg, fecal lipid excretion was 14 kg, and body lipid mass increased 61 kg (Table S4).

At slaughter, lipid allometric coefficients in cows were highest in subcutaneous adipose tissue, followed by perirenal and intermuscular adipose tissue, and the *RA* and *LT* muscles. In calves, this order differed only in that intermuscular adipose tissue had the lowest coefficient (Table S5).

3.2. Input and output mass balance in cows

Control grass silage showed concentrations of 1.6 ± 0.5 μg of iPCBs, 0.07 ± 0.02 ng of dlPCB TEQ and 0.03 ± 0.02 ng of PCDD/F TEQ kg_{DM}^{-1}, whereas exposed grass silage-soil mixture showed concentrations of 6.6 ± 0.8 μg of iPCBs, 0.65 ± 0.15 ng of dlPCB TEQ, and 1.9 ± 0.2 ng of PCDD/F TEQ kg_{DM}^{-1} (Driesen et al., 2022).

The input-output balance for cows is shown in Fig. 2A–D for iPCB and dlPCB TEQ sums, and in Fig. S1 and Table S3 for detailed congeners, including PCDD/Fs. The initial body burden comprised <13% and <3% of the total PCB input (sum of initial burden and dietary intake) in control and treated (exposed and decontaminated) cows, respectively. During the entire experiment, control cows' iPCB and dlPCB TEQ intake was 3.6- and 8.1-fold lower compared to that of exposed cows, and 2.8- and 6.1-fold lower compared to that of decontaminated cows. During the decontamination period (DIM165-288), 4.1-fold and 11.4-fold higher iPCB and dlPCB TEQ intake levels (ng) were recorded in exposed than in decontaminated cows.

In terms of total output (sum of final body burden, calving, milk, and fecal excretion), feces constituted the predominant contribution: 77% of iPCBs, and 40% and 62% of dlPCB TEQ in control and treated cows, respectively (Fig. 2C–D). Milk excretion contributed 13% of iPCBs, and 36% and 27% of dlPCB TEQ in control and treated cows, of which, on average, 69% was ingested by calves. The final burden contributed 13% of iPCBs and 24% of dlPCB TEQ in control cows, 9% and 13% in exposed cows, and 7% and 9% in decontaminated cows, respectively. Calving excretion played a minor role, contributing <0.9%. The estimated metabolized fraction (residuals of the input-output balance) relative to total input was higher in control animals, with 68% for iPCBs and 50% for dlPCB TEQ compared to exposed (29%, 30%) and decontaminated (24%, 23%) animals. Primiparous cows' PCB input and total output were 1.3-fold and 1.2-fold lower, respectively, whereas the final body burden was 1.1-fold higher compared to multiparous cows.

On an individual congener level, output contributions depended on the persistence of the congener. PCB101 (labile example) had a high estimated metabolized fraction (82% for control and 58% for treated animals, relative to input) and fecal excretion (18% for control and 41% for treated animals), whereas milk excretion and body accumulation, represented as final burden, were negligible (contribution of <1%) (Fig. S1). By contrast, the persistent PCB153 showed a low estimated metabolized fraction (<16% for treated animals, relative to input), resulting in body accumulation (10% for control and 9% for treated animals, relative to input), milk excretion (11% for control and 17% for treated animals), or fecal excretion (31% for control and 69% for treated animals).

3.3. Input and output mass balance in calves

The input-output balance for calves is shown in Fig. 2E–H for iPCB and dlPCB TEQ sums, and in Fig. S2 and Table S4 for detailed congeners, including PCDD/Fs. The initial body burden in calves at parturition comprised <4.6% of the total PCB input (sum of initial burden, milk, and solid feed intake). Total iPCB and dlPCB TEQ milk intakes contributed 24% and 51% to the total input of control calves, 77% and 88% to that of exposed calves, and 70% and 83% to that of decontaminated calves. Contribution from solid feed intake increased with lower-chlorinated labile congeners.

Control calves' iPCB intake was 2.3-fold and 1.8-fold lower and dlPCB TEQ intake 3.9-fold and 2.8-fold lower compared to exposed and decontaminated calves, respectively. By design, milk PCB intake differed for exposed and decontaminated calves during DIM165-288, with a 2.3-

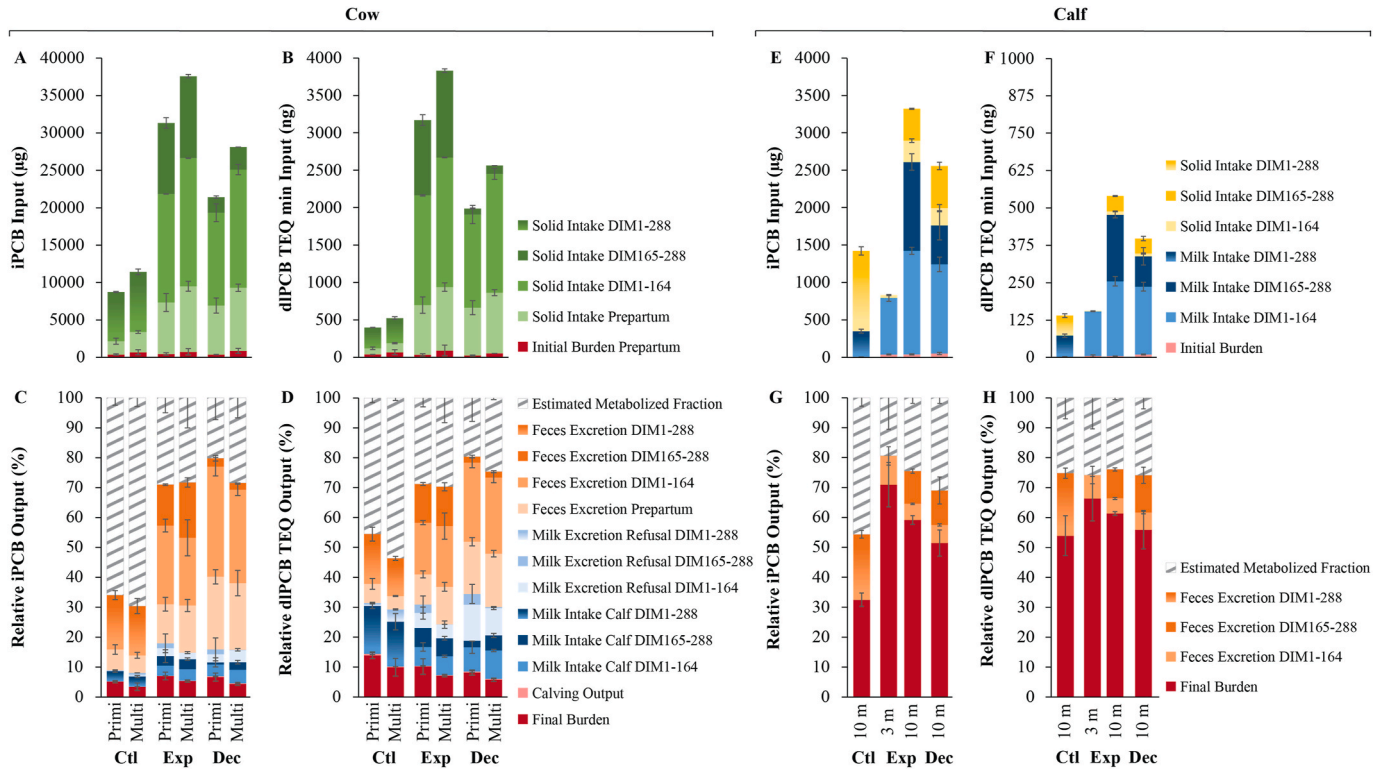

Fig. 2. Input (upper panels) and output (lower panels, normalized to input) mass balance of the sum of iPCBs and dlPCB TEQ min [lower-bound (not detected = 0)] in cows (A–D) and calves (E–H). Cows were divided into primiparous and multiparous animals within each control (Ctl), exposed (Exp), and decontaminated (Dec) treatment ($n = 2$ each). Calves were divided into 10-month-old control ($n = 4$), exposed ($n = 3$), and decontaminated ($n = 3$) and 3-month-old exposed treatments ($n = 2$). The term 'milk excretion refusal' refers to produced milk not ingested by calves. Data related to the decontaminated animals' outputs have to be interpreted with caution due to a switch in exposure level at DIM165 and the representation of relative values. Arithmetic means and standard errors are displayed.

and 2.2-fold higher intake of iPCBs and dlPCB TEQ in exposed calves.

Control calves' PCB output (sum of final body burden and fecal excretion) was 3.5-fold lower compared to that of exposed calves (Fig. 2G–H). The final body burden comprised, on average, 73% of the total PCB output for 10-month-old calves (control, exposed, and decontaminated) and even 89% for 3-month-old exposed calves. This contribution increased with persistence (final burden contributed 5% of PCB101 labile, 74% of PCB114 semi-persistent and 79% of PCB153 persistent congener example to the total output; Fig. S2). Fecal excretion was 1.3-fold higher in exposed calves than in decontaminated calves after DIM164. The estimated metabolized fraction of iPCBs relative to total input was higher in control (46%) than in exposed (25%) and decontaminated calves (31%), whereas similar levels (38%) were found for dlPCB TEQ.

Calf input mass was 9.0-fold lower for iPCBs and 5.1-fold lower for dlPCB TEQ compared to cows, whereas the final body burden in calves and cows was quite similar. Indeed, the final body burden in control and treated calves contributed 3.8- and 8.3-fold more to the total output than in cows.

3.4. Cow absorption rate, assimilation efficiency and metabolism at the congener level

To determine ARs, AEs, and estimated metabolized fractions in cows, individual POP congener mass balances were set up in Fig. 3A–B and Tables S6–7.

Whatever the dietary treatment, the ARs of the tri- and tetra-chlorinated PCBs were between 62% and 83%. For higher-chlorinated PCBs, the exposure treatment had a decreasing effect on AR ($0.08 > P > 0.001$; except for PCB123, $P = 0.90$), which had 1.3-, 1.8-, and 3.0-fold lower ARs for penta-, hexa-, and hepta-PCBs compared to the control. Accordingly, the control group's AR range was 71–87% for penta-chlorinated PCBs, 56–80% for hexa-PCBs, and 42–72% for hepta-PCBs. The exposed group's AR range was 53–78% for penta-PCBs, 36–50% for hexa-PCBs, and 28–42% for hepta-PCBs. The PCB123 was an exception for both groups (85–95%).

Conversely, the exposed cows showed numerically higher ($P < 0.40$) total PCB AEs (sum of milk excretion and body storage) for penta- and higher-chlorinated congeners compared to control cows (except for PCB126 and 189). This resulted in a higher estimated metabolized fraction for control cows compared to exposed cows (similar for PCB28, 52, 77, and 123; 1.4-fold higher for penta- and 3.3-fold higher for hexa- and hepta-PCBs). Sometimes, the PCB AEs had a negative body burden balance component, as mainly multiparous control cows showed body decontamination rather than accumulation during the experiment. When excluding those cases, as well as the tri- and tetra-PCBs from the mean, milk excretion determined 72% of the total AE.

The PCDD/F ARs were lower than those of PCBs, with the highest being 47–54% for TCDD/F, after which AR decreased below 15% for HpCDD/F and OCDD/F. A decreasing effect of the exposed treatment compared to the control treatment could only be confirmed with 1234678HpCDD ($P = 0.04$). Accordingly, PCDD/F total AEs were highest for 23478PeCDF, 2378TCDD, and 12378PeCDD, followed by a decrease as the degree of chlorination increased.

Cows' PCB and PCDD/F ARs and total AEs were not affected by parity ($P > 0.08$; except the AR of 2378TCDF, $P = 0.04$, and 123789HxCDD, $P < 0.001$, and AE of 1234678HpCDF, $P = 0.01$). However, in exposed primiparous cows, ARs were often numerically higher than in multiparous cows. When splitting AE, primiparous cows often had numerically higher net body burden AEs (significant for control PCB126 and 156, $P < 0.05$) and numerically lower milk excretion AEs than multiparous cows.

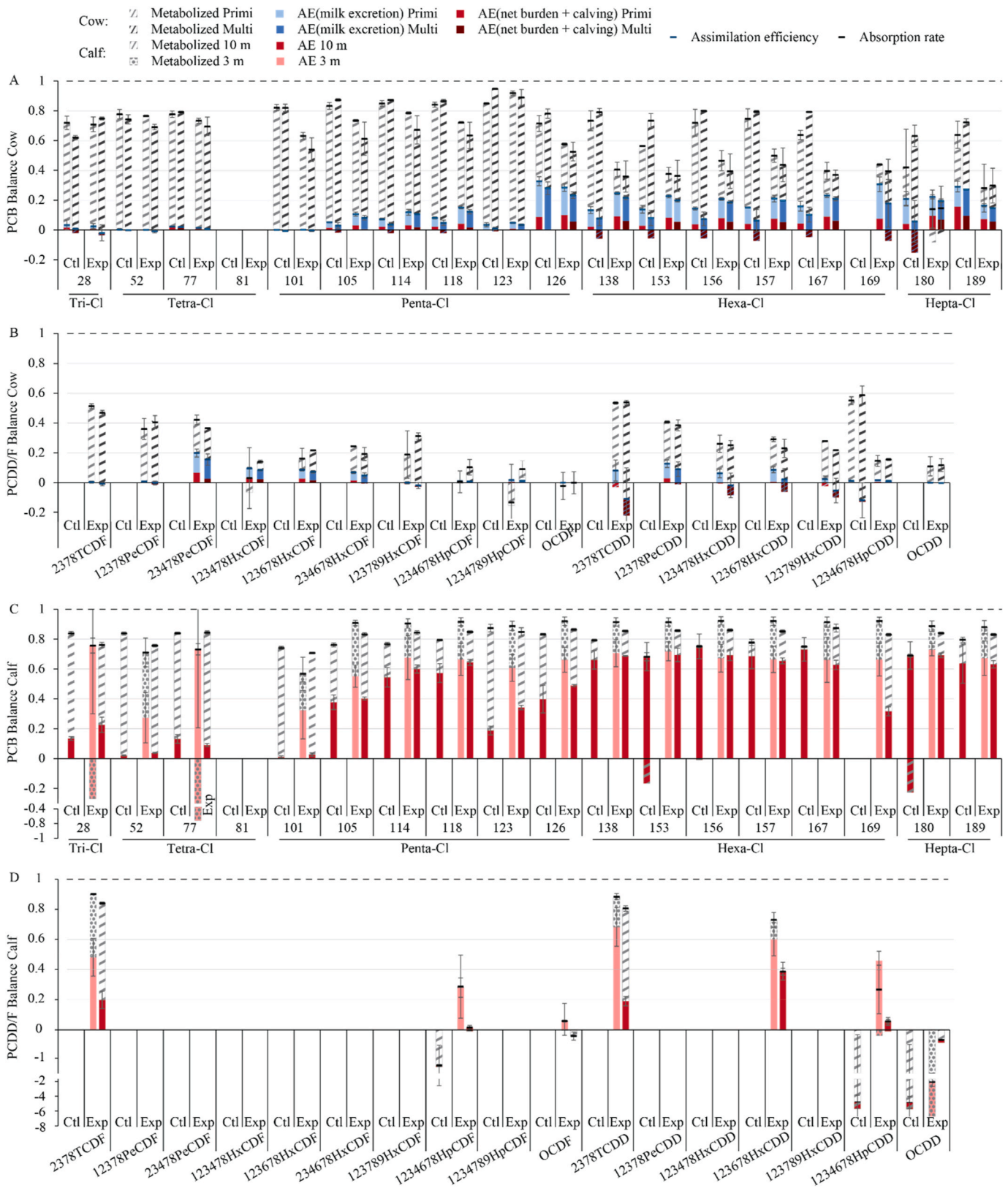

Fig. 3. Absorption rate, assimilation efficiency (AE), and residual (assumed to be metabolized) fractions for cows (A, B) and calves (C, D) of each individual PCB (A, C) and PCDD/F (B, D) congener detected throughout the matrices. The cows' AEs were further subdivided into body tissue storage (net body burden + calf at parturition) and milk excretion fractions. Negative values for net body burden related to decontamination processes throughout the experiment. Means composed of $n = 2$ for cows in each treatment × parity group, $n = 4$ for control calves, $n = 2$ for 3-month-old exposed calves, and $n = 3$ for 10-month-old exposed calves. Arithmetic means and standard errors are displayed.

3.5. Calf absorption rate, assimilation efficiency and metabolism at the congener level

The individual POP congener mass balances used to determine ARs, AEs, and estimated metabolized fractions for calves are presented in Fig. 3C–D and Tables S8–9.

Unlike in cows, calves' PCB ARs were not influenced by the degree of chlorination and showed similar ARs for PCB105 and higher-chlorinated PCB congeners. The ARs in exposed 10-month-old calves were, on average, 2-fold higher than in exposed cows. For PCB28, 52, 77, and 101, PCB ARs were higher in 10-month-old control calves than in exposed calves (81% vs. 77% for 10-month-old exposed calves, with PCB28 $P = 0.04$, and 69% for 3-month-old exposed calves, $P < 0.05$). Conversely for higher-chlorinated PCB congeners, ARs were lower in control calves (77%) than in 10-month-old exposed calves (85%, $P < 0.04$, except PCB123, 126, and 189) and 3-month-old exposed calves (91%, $P < 0.05$).

Overall, the calves' PCB body burden AEs were higher than those of the cows' total AEs and did not differ between dietary treatments, except for PCB28, 52, 101, 123, and 169. In these latter cases, AEs were lowest ($P < 0.05$) in the control group, followed by 10-month-old and then 3-month-old exposed calves. The estimated metabolized fraction mainly decreased as the degree of chlorination increased. For the lower-chlorinated congeners (PCB28, 52, 77, 101, 123), control calves had the highest estimated metabolized fraction (73%), followed by 10-month-old (64%) and 3-month-old exposed calves (32%). For higher chlorination, smaller values and no treatment effect were recorded (on average, 24% when excluding control PCB156 and 167).

For calf PCDD/F ARs, a numerical decrease with increasing chlorination still occurred. The few calf PCDD/F AEs determined were higher in 3-month-old than in 10-month-old exposed calves, and both were higher than those in cows. The estimated metabolized fractions were highest for TCDD/F.

3.6. Tissue distribution in cows and calves

Cows' and calves' empty body (full body without digesta and urine) iPCB and dlPCB + PCDD/F TEQ concentrations and POP distribution among tissues at slaughter (DIM288) are displayed in Fig. 4 for sums and in Figs. S3–4 and Tables S10–11 for detailed congeners.

3.6.1. Empty body concentrations in cows and calves

Cows' empty body POP concentrations were highest in exposed cows, with 24 ng of iPCBs and 6.6 pg of dlPCB + PCDD/F TEQ g^{-1} lipids – 1.7- and 2.4-fold higher than in decontaminated cows ($P = 0.11$ and $P < 0.001$) and 6.5- and 12.5-fold higher than in control cows ($P = 0.002$ and $P < 0.001$), respectively. Empty body iPCB and dlPCB + PCDD/F TEQ concentrations in primiparous cows were numerically ($P > 0.11$)

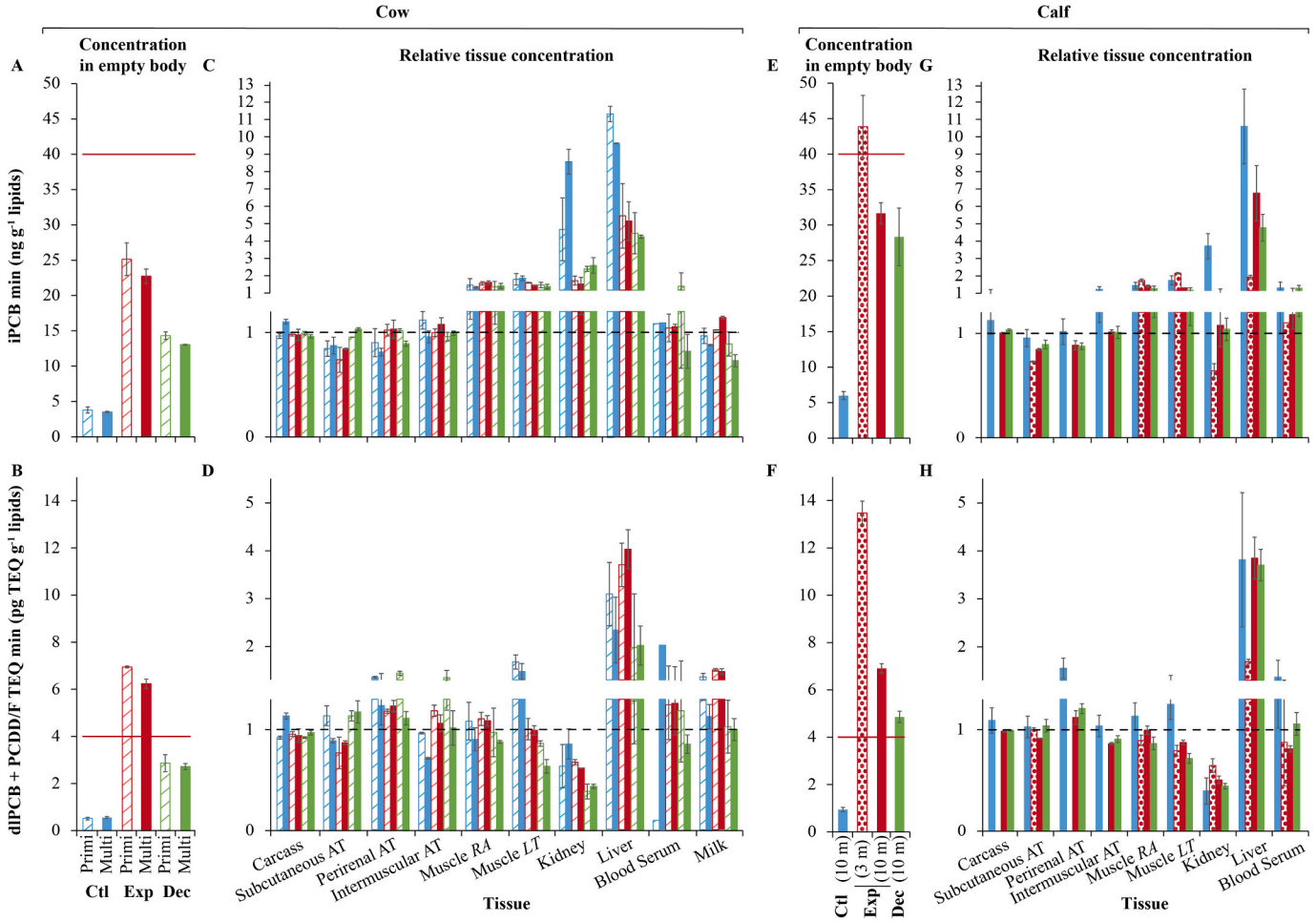

Fig. 4. Empty body concentrations for the sum of the iPCBs (upper panels) and dlPCBs + PCDD/Fs (lower panels) in cows (A–B) and calves (E–F), as well as concentrations in different tissues normalized to the empty body in cows (C–D) and calves (G–H), are represented. The red horizontal line represents the EU maximum regulatory level in bovine meat or adipose tissue [Regulation (EU) Nr. 1259/2011]. Means composed of $n = 2$ for cows in each treatment × parity groups, $n = 4$ for control (Ctl) calves, $n = 2$ for 3-month-old exposed (Exp) and $n = 3$ for 10-month-old exposed and decontaminated (Dec) calves. Arithmetic means and standard errors are displayed. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

1.1-fold higher than in multiparous cows, except for control cows' concentrations of dlPCB + PCDD/F TEQ. Among calves, empty body iPCB and dlPCB + PCDD/F TEQ concentrations were highest in 3-month-old exposed calves, having 44 ng and 13 pg TEQ g^{-1} lipids – 1.4- and 2.0- fold higher than their 10-month-old counterparts ($P = 0.41$ and $P < 0.001$), 1.5- and 2.8-fold higher than in decontaminated calves ($P = 0.30$ and $P < 0.001$), and 7.3- and 14-fold higher than in control calves ($P = 0.009$ and $P < 0.001$), respectively. When comparing cows and calves, empty body iPCBs and dlPCB + PCDD/F TEQ concentrations were 1.3- and 1.1-fold higher in exposed calves, 2.1- and 1.7-fold higher in decontaminated calves, and 1.6- and 1.8-fold higher in control calves compared to the respective cows.

3.6.2. Tissue distribution based on sums

Neither the effects of dietary treatment in cows and calves, nor parity in cows, on POP distribution among tissues were observed, when normalized to the empty body concentration, except for the liver ($P < 0.001$) and kidneys (only for iPCBs in cows, $P < 0.001$).

In particular, the liver (in all cows and 10-month-old calves, $P < 0.001$) and, to a lesser extent, the kidneys (in all cows and control calves, $P < 0.01$) showed significantly higher iPCB concentrations than other tissues when normalized to the empty body, with only exposed cows' kidneys having significantly higher relative concentrations than subcutaneous adipose tissue ($P = 0.03$). Liver and kidney iPCBs of 3-month-old calves were an exception, with liver levels comparable to those in muscles and numerically lower kidney levels compared to the empty body ($P = 0.73$). Besides offal, cows' and calves' *RA* and *LT* muscles showed numerically higher iPCB concentrations than other tissues (significant for control cows' *LT* iPCBs, $P < 0.04$ vs. all tissues, except serum, $P = 0.08$). Slightly lower values of iPCB concentrations in the subcutaneous adipose tissue of control and exposed cows (vs. control cows' *LT*, $P = 0.01$, and exposed cows' *LT*, $P = 0.06$, and *RA*, $P = 0.05$) and exposed and decontaminated calves were recorded. In addition, iPCB levels in perirenal adipose tissue were numerically lower in control cows (vs. *LT*, $P = 0.01$) and exposed and decontaminated calves. Carcass, adipose tissues, blood serum, and milk had similar iPCB concentrations to those found in empty bodies of cows and calves.

Liver dlPCB + PCDD/F TEQ concentrations were also higher than those in other tissues ($P < 0.02$; 3-month-old exposed calves, $P = 0.08$), whereas cows' and calves' kidney concentrations showed lower levels. Cows' perirenal and intermuscular adipose tissue, blood serum, and milk had numerically higher dlPCB + PCDD/F TEQ concentrations than the empty body (exposed milk vs. subcutaneous adipose tissue, $P = 0.04$), whereas in calves, concentrations in intermuscular adipose tissue, muscle *LT*, and blood were slightly lower.

3.6.3. Tissue distribution based on congener patterns

Congener patterns in cow and calf tissues showed slight iPCB variations (Tables S12–15). The hexa-chlorinated PCB138 (35% of iPCB sum in cows and 36% in calves) and 153 (40% and 44%) were mostly represented in the empty body, carcass, adipose tissues, liver (of calves), blood serum, and milk, whereas in the muscles and livers of treated (exposed and decontaminated) cows, PCB52 (18%) was also represented. In cows' and calves' kidneys and control cow's livers, the main contribution shifted to PCB52 (36% in cows and 28% in calves) and 101 (30% and 25%). Among the dlPCBs (raw concentrations), PCB 118 (62% and 66%) dominated all tissues, followed by PCB105 (14% and 11%) and 156 (11%). Among PCDD/Fs in the tissues of treated cows and calves, generally, 123478HxCDF (22% and 31%) was found next to 1234678HpCDF (15%) in cows, and 23478PeCDF (10%) or 123678HxCDF (12%) in calves. In exposed animals, the contribution of OCDD was also important for cows' kidneys and livers (30%) and primiparous cows' muscles (22%), whereas for calves, an average contribution of 23% was seen throughout the tissues.

4. Discussion

The study of PCB and PCDD/F fate in the present transgenerational setting includes the understanding of pollutant ADME processes under the influence of lipid dynamics in reproductive cows and growing calves. This study successfully mimicked a 'classical' suckling cow husbandry (Driesen et al., 2022), which made it feasible to investigate ADME processes throughout gestation and the entire lactation period for such extensive beef production systems based on a complete cow-calf mass balance and tissue distribution approach.

4.1. ADME of PCBs and PCDD/Fs in lactating cow

Following ingestion, POPs are absorbed from the intestine to the blood and lymphatic system, which act as distribution compartments (Amutova et al., 2020). Apparent AR, calculated via the intake-fecal excretion balance (Kierkegaard et al., 2009; McLachlan, 1993; Thomas et al., 1999), was shown to be influenced by the exposure matrix (e.g., oil, forage, and soil), POP physicochemical characteristics, and digestive physiology (Amutova et al., 2020). The POP exposure matrix (grass silage for control and mainly soil for exposed cows), with a potential additional effect coming from the concentration difference, widely affected AR, which in exposed cows was progressively reduced from penta- until hepta- and octa-chlorinated PCBs and PCDD/Fs compared to control cows. This discrepancy among treatments was presumably due to a reduced bioaccessibility of highly chlorinated POPs bound to soil compared to those provided 'freely' by the control grass silage (POP origin in the latter case was assumed to be atmospheric deposition rather than soil particles based on levels of POPs and soil markers recorded in the control grass silage and grassland soils where it was harvested). Conversely, Richter and McLachlan (2001) found no differences between the ARs of POPs from atmospheric deposition and the soiling of grass forage. Besides, higher chlorine numbers are linked to a higher lipophilicity (log K_{OW}). This might hamper, at some point (log $K_{OW} >$ 6.5), POP absorption across the intestinal wall (McLachlan, 1993) due to the mechanistic concept of having to cross the water layer surrounding intestinal microvillosities (Kelly et al., 2004). Nonetheless, when plotting AR against log K_{OW}, the PCB and PCDD/F ARs in control cows were less affected by the degree of chlorination than in exposed cows (Fig. 5). A model describing ARs from another dairy cow study (Kelly et al., 2004; McLachlan, 1993) is also plotted in Fig. 5, highlighting its consistency with the present data, as pasture background exposure in McLachlan (1993) resulted in ARs between the control and exposed treatments ARs

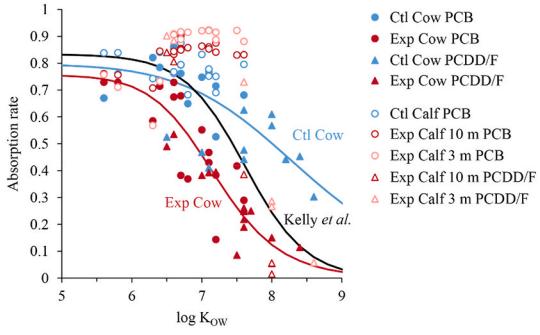

Fig. 5. Apparent dietary absorption rates (ARs) of PCBs ● and PCDD/Fs ▲ in cows and calves (◯, Δ) against their chemical log K_{OW} (Amutova et al., 2020). Trend lines represent a Hill function fitted for control (Ctl, $n = 4$) cows (blue): $AR = (0.77 \times \log K_{OW}^{-9.38})/(8.79^{-9.38} + \log K_{OW}^{-9.38})$; a Hill function for exposed (Exp, $n = 4$) cows (red): $AR = (0.75 \times \log K_{OW}^{19.03})/(7.23^{-19.03} + \log K_{OW}^{19.03})$; and a non-linear regression for dairy cows (black) (Kelly et al., 2004): $AR = 1/(2.9 \times 10^{-8} \times K_{OW} + 1.2)$. A relationship for calves could not be fitted. Negative apparent ARs for specific congeners were not represented. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

calculated here.

Multiparous exposed cows had a numerically 1.1-fold lower PCB AR compared to primiparous cows, which is in line with the higher lipid digestibility in primiparous (1.1-fold higher in primi- vs. multiparous cows). This results in an increased POP fecal output flux in multiparous animals, which consequently decreases the AR, as established in the fugacity digestive model of Kelly et al. (2004).

The actual POP fraction assimilated into cows was determined via the computation of AE, which was further divided into a milk excretion part and a body accumulation part (net cow body burden and calf excretion at parturition). Cows' PCB total AEs increased, whereas the PCDD/F AEs decreased with chlorination degree, except for the low AEs of TCDF and 12378PeCDF. The same trends have previously been reported for transfer rates to milk (Driesen et al., 2022; Hoogenboom et al., 2015a). Although PCB and PCDD/F total AEs were not significantly affected by parity, primiparous cows tended to have higher net body burden AEs and lower milk AEs, compared to multiparous cows. This is in accordance with the lower daily transfer rates to milk at DIM164 in primiparous cows (Driesen et al., 2022). The increased body AE in primiparous cows resulted from a reallocation of the POP flux from milk excretion to body storage, not only because the milk fat yield was 1.9-fold lower in primiparous cows, but also because they were still growing (estimated body lipid mass increased 3.6-fold more during the study in primi- vs. multiparous cows). Therefore, primiparous cows were in an anabolic lipid accretion stage that drove POPs to storage compartments (net body burden 1.9-fold higher in primi- vs. multiparous cows) rather than to mammary glands for milk excretion.

The estimated metabolized fractions confirm the previous classification of congeners as labile (PCB28, 52, 77, 101, and 123), semi-persistent (PCB105, 114, and 118), and persistent (PCB126, 138, 153, 156, 157, 167, 169, 180, and 189) (Driesen et al., 2022). McLachlan (1993) previously linked PCB persistence to the 4,4′ substitution pattern, but the present study shows that this cannot be the only explanation in light of the high estimated metabolized fraction of 28, 77, and 123, for example.

Higher estimated metabolized fractions for penta- and higher-chlorinated PCBs in control compared to exposed cows might be explained by a saturation of the hepatic enzyme system in the latter. Unfortunately, no hepatic enzyme activity measurements were performed to confirm such an assumption. Alternatively, this may have resulted from body decontamination during the experiment seen in the control vs. the accumulation stage in exposed cows. More POPs could then reach the liver of control cows when expressed relative to the total body burden. Conversely, the accumulation process in exposed cows probably resulted in the allocation and subsequent storage of POPs in the adipose tissue sink, further limiting their access to the liver for metabolization.

In terms of control and treated cows' total outputs, feces constituted the predominant excretion route, contributing 77% of iPCBs and 40% and 62% of dlPCB TEQ, which increased with the degree of chlorination (Fries et al., 2002; McLachlan and Richter, 1998). The fecal excretion of HpCDF and OCDF was even in excess of intake, which might be attributed to synthesis by micro-organisms in cows' digestive tracts. This was assumed previously, but based on an excess of HpCDD and OCDD (Fries et al., 2002).

4.2. ADME of PCBs and PCDD/Fs in suckling calves

Calf PCB input starts during prenatal exposure, since the placental barrier seems ineffective against such compounds (Hirako et al., 2005). Nonetheless, the initial body burden at parturition of 10-month-old calves contributed <2.5% to the total input. Therefore, the cow-calf transgenerational transfer of POPs occurs mainly *postpartum* through suckling (Hirako, 2008).

Unlike cows, calves' PCB ARs did not decrease with increasing chlorination (Fig. 5), which is in accordance with the findings of Hirako (2008). One explanation could be the higher lipid digestibility observed in calves (87%) compared to cows (63%), as well as the 2-fold higher body lipid accretion. The latter potentially promoted POP body dilution and subsequently resulted in a higher POP concentration gradient between digestive content and the body. Moreover, the absorption of persistent POPs, previously bioavailable in cows, might also be promoted by the richer content of highly bioavailable lipids (i.e., triglycerides) in milk compared to grass silage or soil. Thus, the steady AR levels of pre-weaning (i.e., pre-ruminant) calves, even for the highly chlorinated PCBs that were tested, coincided with observations in birds and humans, both having a high dietary lipid content and digestibility, and POP ARs that stayed high (>80%) until reaching a log K_{OW} of 8 (Kelly et al., 2004). Accordingly, calf 1234678HpCDD/F and OCDD/F ARs showing a log $K_{OW} \geq 8$ decreased, as observed by Hirako (2008).

Whatever the treatment, the calves' net body burden AEs were higher than the total AEs in cows, which mirrors the higher ARs in calves compared to cows.

As in cows, the estimated metabolized fraction decreased with the degree of chlorination in calves. The 3-month-old calves showed the lowest estimated metabolized fraction. This is corroborated by the lower relative POP concentrations seen in the liver, which subsequently results in less cytochrome binding and therefore reduced metabolization by the hepatic enzymes in such young and non-mature cattle. Similarly, Scheuplein et al. (2002) mentioned that human infants younger than 6 months old lack the capacity to metabolize contaminants as readily as adults.

4.3. Tissue distributional fate of PCBs and PCDD/Fs in cows and calves

Empty body iPCB and dlPCB + PCDD/F TEQ concentrations were highest in 3-month-old calves, followed by 10-month-old calves and then cows, which resulted at least in part from calves' lower BW and fatness (Driesen et al., 2022). The same effect was seen in cows, with higher empty body concentrations for primiparous cows, which is consistent with the higher net body burden AEs and previously determined milk concentrations (Driesen et al., 2022). Besides determining the actual body burden, investigation of tissue distribution is necessary to identify where the compounds specifically accumulate, especially in tissues and organs that play key roles in ADME (i.e., blood, adipose tissue, liver, and milk) or that are edible and thus entail further implications for food safety (i.e., muscles, offal, and milk). Relative to other tissues, a clear enrichment of iPCBs and dlPCB + PCDD/F TEQ was found in the livers of cows and calves, which might result from specific hepatic cytochrome P450-binding, an enzyme involved in the hydroxylation and thus metabolization of these POPs (Inui et al., 2014). Such enrichments have already been reported in lactating cows (Richter and McLachlan, 2001; Thomas et al., 1999) and especially in sheep (Hoogenboom et al., 2015b; Lerch et al., 2020). This POP enrichment in the liver was less pronounced in 3-month-old calves, which is in accordance with their lower estimated metabolized fraction. In kidneys, PCB enrichment was less pronounced and congener-specific. As for PCDD/Fs (hexa, hepta, octa), only higher levels were found in the control kidneys. These results suggest that similar protein binding as in the liver occurs in the kidneys of mature cattle. This assumption is corroborated by 3-month-old calves showing lower kidney concentrations in which, e.g., iPCBs were dominated by PCB138 and 153 instead of PCB52 and 101 in older calves.

When comparing muscles and adipose tissue, iPCB concentrations (mainly PCB28, 52, and 101) were highest in *LT* > *RA* > intermuscular > perirenal > subcutaneous adipose tissue, following the exact order of increasing lipid allometric coefficients (Table S5) (Chilliard et al., 1998). An increase in body fat mass, as seen throughout the last months of the experiment, resulted in a higher rate of lipid deposition within subcutaneous adipose tissue and thus a relatively higher POP dilution and lower concentration than in muscles in which lipid deposition and subsequent POP dilution occur at a slower rate. Additionally, compared

to muscles, adipose tissues are deep compartments that are poorly perfused by blood and are characterized by permeability-limited fluxes (MacLachlan, 2009). This results in an equilibrium delay with the blood compartment and, consequently, other tissues, further enhancing the dilution effect due to lipid deposition. In accordance but reversely, a POP concentration gradient in the respective order perirenal > subcutaneous > mesenteric adipose tissue > *RA* muscle corresponding to decreasing lipid allometric coefficients occurred over a period of body lipid mobilization in underfed ewes (Lerch et al., 2020). Besides, in the present study the altered iPCB pattern, with high contributions from lower chlorinated PCBs in *LT* and *RA* (52 and 101 vs. 138 and 153 in other tissues), might suggest additional protein binding in muscles, thus promoting enhanced enrichment.

For dlPCB + PCDD/F TEQ, such an allometric trend was identified in exposed cows for PCB77, 81, 105, 114, and 123, but to a much lesser degree. In calves, this trend was seen for PCB77 and 81, as well as in exposed calves for 123478HxCDD/F, 12378HxCDD/F, 1234789HpCDF, and OCDD.

The decontamination process was tissue-specific, which might also be important to consider in risk assessment, as the largest concentration differences between exposed and decontaminated animals were seen in high turnover (milk) and well-perfused tissues (liver and muscles). Overall, the fold decrease in concentration among decontaminated and exposed animals was larger in cows than in calves. The switch to control grass silage for cows led to an abrupt decrease in exposure level per kg BW, whereas milk continued to deliver POPs to decontaminated calves. These results might seem controversial in light of previous observations in which a stronger decontamination potential in the subcutaneous adipose tissue of calves compared to cows was found between DIM164 and DIM288 (Driesen et al., 2022). However, this smaller empty body POP concentration difference between exposed and decontaminated calves at DIM288 might have resulted from the additional dilution effect seen in exposed calves with a higher rate of body lipid deposition compared to cows.

Based on these results and the fact that the liver contributed only 0.3% to the total burden, it might be possible to estimate the total body burden by multiplying the lipid-based concentration of one adipose tissue depot by an estimate of empty body lipid mass (Richter and McLachlan, 2001). To achieve a reliable estimate with a low influence from physiological dynamics, intermuscular adipose tissue would be the most suitable due to its lipid allometric coefficient being the closest to one and, accordingly, its best aligned POP concentration with the entire empty body. Muscle tissue itself could lead to an overestimation of empty body concentrations due to its allometric coefficient of less than one and because the distribution there seems to not only be limited to lipid dynamics. Conversely, subcutaneous (*in vivo* monitoring) and perirenal (easy sampling without depreciation of the carcass for *post-mortem* monitoring) adipose tissue levels might not totally reflect muscle (i.e., meat) concentrations, as for iPCBs and dlPCB + PCDD/F TEQ, differences up to 2-fold were identified.

5. Conclusion

The results of this study showed the overall transgenerational ADME fate of PCBs and PCDD/Fs in cows and calves exposed via a grass silage-soil mixture. Absorption rates and AEs were shown to be affected by the physicochemical properties of POPs, the exposure matrix, and animal physiology. The AR of PCBs and PCDD/Fs in cows decreased with an increasing chlorination degree. An additional decrease for highly chlorinated compounds was recorded in soil-exposed cows compared to control cows, presumably due to reduced bioaccessibility when POPs were carried through soil vs. grass silage. In calves mainly exposed to POPs via milk, PCB ARs were high (69%) and not influenced by chlorination, with values close to those recorded previously in monogastric (e.g., humans). This highlights the importance of considering the exposure matrix when handling ARs for risk assessment. Unlike cows, calves' fecal excretion was low, and they mainly accumulated the POPs in their bodies. This highlights the importance of lipid deposition during growth and subsequent POP dilution to reduce meat contamination levels in suckling calves. When chlorination was increased, the AE increased for PCBs, whereas it decreased for PCDD/Fs. The AE was dependent on the physiological status of the cattle (i.e., lactating or growing, primiparous or multiparous), further challenging the previously assumed independence of AE from physiological traits. When compared to multiparous cows, the milk and net body burden AEs in primiparous cows were lower and higher, respectively, presumably due to a lower milk fat yield but higher body lipid accretion. Tissue distribution reveals a specific accumulation in the liver, followed by the kidneys and muscles for some congeners. The POP sum concentrations in adipose tissue (especially intermuscular) were the closest to that of the entire empty body and may be further used to estimate the body burden, in case a reliable estimate of body lipid mass is available. Nonetheless, adipose tissue concentrations underestimated levels in the most consumed beef tissues (i.e., muscles) of cows and calves by up to 2-fold, according to tissue lipid allometry. This specific trend warrants particular attention in risk assessment and monitoring. Therefore, to ensure chemical food safety in suckling beef husbandry, it is important not only to focus on POP physicochemical properties, but also to take into account body lipid dynamics and the exposure matrix. Physiologically-based toxicokinetic (PBTK) models are useful for handling and integrating the complexity of the ADME-physiology interplay and the dataset provided here will facilitate the further calibration and validation of fine POP PBTK models (Bogdal et al., 2017; Lerch et al., 2018).

Credit author statement

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The authors declare no competing financial interests.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

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